TRANSACTIONS of the BOSE RESEARCH INSTITUTE CALCUTTA

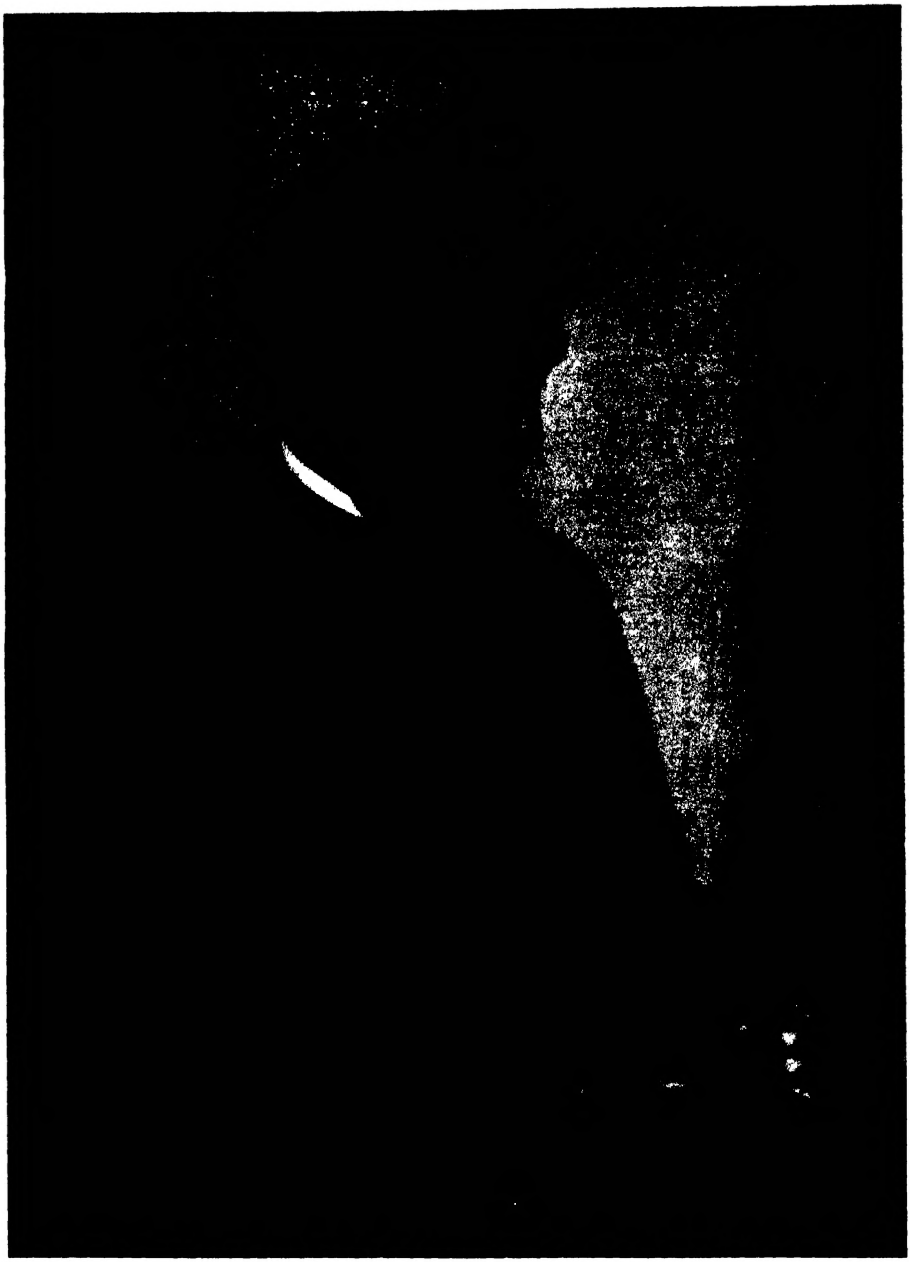
STARTED IN 1918 BY SIR J. C. BOSE

Vol. XII, 1936-1937

Edited by
D. M. BOSE, M.A., PH.D., F.N.I.
Director, Bose Research Institute

THE BOSE RESEARCH INSTITUTE
93, UPPER CIRCULAR ROAD, CALCUTTA

CONTENTS



Jagadish Chandra Bose

30th November 1858 - 23rd November 1937

SIR JAGADISH CHANDRA BOSE MEMORIAL ADDRESS

By DR. RABINDRA NATH TAGORE

30th November, 1938

When by some fortunate chance I came into an intimate contact with Sir Jagadish, he was in the prime of his youth and I was very nearly of his age. At that moment his mind seemed entranced with a vision of the living creatures' fundamental kinship with the world of the unconscious. He was busy in employing his marvellous inventiveness in coaxing mute Nature to yield her hidden language. The response which he received through skilful questionings revealed to him glimpses of the mystery of an existence that concealed its meaning underneath a contradiction of its appearance. I had the rare privilege of sharing the daily delight of his constant surprises. I believe, poets inherit the primeval age in their temperament when things in their infant simplicity revealed a common feature. Somehow these lovers of *Maya* feel the joy of their being spread all over the creation, which makes them indulge in seeking the analogy of the living in things that appear lifeless. Such an attitude of mind may not in all cases be based upon any definite belief, animistic or pantheistic; it may be merely a make-believe, as we notice in children's play, which owes its origin to the lurking tendency in our sub-conscious mind to ascribe life-energy to all activities in the natural world. I was made familiar from my boyhood with the Upanishad which, in its primitive intuition, proclaims that whatever there is in this world vibrates with life, the life that is one in the infinite.

This might have been the reason of the eager enthusiasm with which I expected that the idea of the boundless community of life in the world was on the verge of a final sanction from the logic of scientific verification. Being allowed to follow the Master's footsteps in the privacy of his pursuit, even though as a mere picker of his casual hints, I had my daily feast of

wonders. At this early stage of his adventure when obstacles were powerfully numerous and jealousy largely predominated over appreciation, friendly companionship and sympathy must have had some needful value for him even from one who to maintain intellectual communion with him lacked special competency. Yet I can proudly claim to have helped him in some of his immediate needs and occasional hours of despondency in those days of an inadequate recognition and feeble support that he received from the public.

In the background of that distant memory of mine, I find not the slightest gleam of a vision of the enormous success that could before long combine scientific renown with a vast material means adequate enough to build this Institute, one of the very few richly endowed mediums in India for bestowing the benediction of science upon his countrymen. In fact, it makes me laugh at myself today to read, in some of my old letters, my effort to encourage him with the likelihood of filling the gaps in his funds when my own resources were precariously limited to persuading friends who were foolish enough to have faith in me. Still it is comically sweet to think of the proud magnificence in my assurance fitfully accompanied by contribution absurdly poor compared to the ceaseless flow of tribute that, later on, he could attract by his own magnetic personality and also by the general confidence he widely aroused in his genius. But I repeat again, it was sweet to have dreamed impracticable dreams and to have done however little it was possible, as it proves a courage of joy in the faith in greatness which itself is a bounteous gift to one's own mind.

However ill equipped as I was by the deficiency in my training and by the poet's idiosyncrasy to be a fit companion to a man of science at a luminous period of his self-revelation, I was still accepted as his close friend and, possibly because of the contrariety in our natural vocations, I was able to offer some stimulation to his urge of fulfilment. Not having the necessary amount of vanity in my constitution, it had been the subject of constant wonder in my mind.

Since then time passed quickly, maturing the fruits of our expectation. During this period of his fast growing triumph,

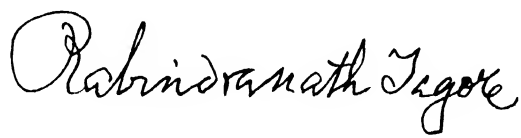
I was modest enough to feel less and less the urgency of my comradeship in his journey towards the goal, which was no longer arduous or beset with uncertainty. And yet I can rightfully claim the credit for strengthening in some measure his trust in his own destiny, by adding to it my own unwavering faith, at that painfully hesitant moment of fortune during the dubious dawn of his career, when even persons of meagre resources might have some important use.

Victory is the inalienable claim of all genuine power having the might of attraction that naturally exploits all kindred elements on its path and moulds them into an image of glory. And such an image is this Institute, which represents the Master's lifelong endeavour taking a permanent shape in the form of a centre for the inspiration of similar endeavours.

However, the early association of mine with the Master's first great challenge of genius to his fate, whose path at that time did not run smooth, belongs for me to a remote period of a history in which I feel myself hazily indistinct. And this made me seriously waver to accept the invitation for taking an honoured seat at a ceremonial meeting in this institution. The presumptuousness of youth made me absurdly proud to imagine that my companionship was growing into an organic part in the history that was being evolved before my eyes, and, in that belief I did try to hearten the hero, which was a part of my vanity. But foolish youth does not last for ever, and I have had time to come to realize my limitation. Anyhow it is quite obvious, that I am a mere poet carrying on my *sadhana* in the temple of language, the most capricious deity who is apt to ignore her responsibility to logic, often losing herself in the nebulous region of fantasy. Our oriental custom is to bring proper gifts to sacred shrines, but my gift of words for this occasion cannot but be out of place among the records of memorable proceedings of a learned society.

Fortunately there are some few men among us who can claim fellowship with the aristocracy in the realm of science, and can be expected to make splendid this ceremony with the wealth of their thoughts. I can only bless this institution from that obscure distance where the multitude of the uncared-for

generations of this country have helplessly drifted to the pitiless toil of primitive land-tilling. I offer my salutation to the illustrious founder of this Institute, humbly sitting by those who are deprived of a sufficiency of that knowledge which only can save them from the desolating menace of scientific devilry and from the continual drainage of the resources of life, and I appeal to this Institute to bring our call to science herself to rescue the world from the clutches of the marauders who betray her noble mission into an unmitigated savagery.



I. EFFECT OF REDUCED ATMOSPHERIC PRESSURE ON THE GROWTH OF PLANTS.

By B. K. DUTT and A. GUHA-THAKURTA.

(*Received for publication 15th August, 1938.*)

In a previous communication [1] the effect of low atmospheric pressure on the pulsatory activity and on the moto-excitability of plants had been most exhaustively reported. It has been shown in that paper that the changes in the pulsatory activity and also on the moto-excitability of plants in reduced pressure is only due to reduction of the partial pressure of oxygen and not due to the reduction of the total pressure as such. Further investigation on this line was continued in studying the effect of reduced pressure on the growth of plants.

Regarding the effect of reduced pressure on growth of plants a few stray observations have been recorded by some workers.

Whitfield [2] showed from a series of observations on the growth of plants in the plains, montane and alpine stations that the greatest increase took place in the plains and least in the alpine station. Further, observing the alpine vegetations composed of dwarf grasses and sedges he remarked that this is probably due to the reduced air pressure which expresses itself most strongly in the low air and soil temperature.

Shope Paul [3] observed in *Populus tremuloides* collected at 5,000 ft. and 9,000 ft. that there was no difference in size and anatomy of leaves at the different altitudes but the twigs are lengthier and thicker at the lower altitude.

Wilson [4] in connection with his studies on the effect of partial pressures of various gases, on the nitrogen fixation process in the inoculated leguminous plants made some observations on the effects of atmospheric pressures on growing red clover plants. He observed that at a pressure below 0·20 atmosphere there was inhibition of growth and remarked that the inhibition was probably due to the low pressure of oxygen.

From the above literatures it will be evident that the different investigators observed an inhibition of growth in reduced atmospheric pressure. But Wilson remarked that the inhibition is probably due to the reduction of partial pressure of oxygen. We, therefore, thought it worth while investigating more thoroughly the effect of reduced atmospheric pressure on the growth of plants and find out definitely whether that effect is due to the reduction of partial pressure of oxygen present in the atmosphere.

In the present investigation on growth of plants, observations were made by recording the effects of different barometric pressures artificially produced in a glass chamber. Experiments were separately performed on shoots and on roots of plants.

Method of Experiment in Studying the Effect of Reduced Pressure on the Growth of Shoots.

For producing low barometric pressure a vacuum desiccator and an electrically driven rotary pump were employed; the detailed description of the method has been given in our previous communication.[1]

The growth was recorded in a single Lever Oscillating Plate Recorder. The plant was vertically attached to the lever. The magnification was 100 times in all the experiments. The lateral movement of the recording plate lasted for 75 minutes which was the maximum time-limit of each experiment. The dots in the records were at intervals of 30 seconds.

The experiments were performed with intact potted seedlings of *Cajanus* and *Helianthus*. The plant was tightly clamped at the base of the stem, the tip being attached to the lever by a silk thread. The experimental arrangement is diagrammatically represented in figure 1.

The pressure was lowered to a certain definite value after the growth had been recorded in the normal pressure for a certain period. When the effect of a particular pressure was recorded for a considerable time, the pressure was brought back to normal in order to record the process of recovery. In this way the effects of reduced pressure from 100 mm. to 500 mm. below normal were repeatedly observed in the above plants.

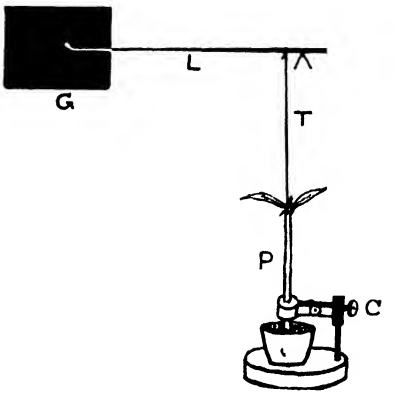


FIG. 1. Diagrammatic representation of recording growth of shoot.
P, plant, clamped at C and attached by the silk thread T, to the lever of the recorder L. G, smoked glass plate for recording growth of the plant.

In seedlings of *Cajanus* the growth rate was found to remain unchanged up to 300 mm. below normal within the time-limit of our observation, but at 400 mm. below normal the growth rate became appreciably decreased. Typical records of the effects of variation of pressure on growth of *Cajanus* are given in figures 3 and 4 (Plate 1). Reduction of pressure is marked with an arrow; an arrow within circle indicates bringing back to normal pressure. Below is given the description of typical experiments.

At 360 mm. or 400 mm. below normal (Fig. 3, Plate 1), the growth rate at arrow is seen to be greatly diminished. When the normal pressure was restored the growth attained the former value.

At 260 mm. or 500 mm. below normal (Fig. 4, Plate 1), the growth after reduction of pressure was almost stopped; after restoration to normal pressure there was gradual recovery.

The growth of *Helianthus* was found to remain unaffected even at a pressure of 360 mm. or 400 mm. below normal within the time-limit of our observation. At 260 mm. or 500 mm. below normal, however, the growth was appreciably decreased. A typical record of the decrease of growth of *Helianthus* in reduced pressure is given in figure 5 (Plate 1).

At 260 mm. or 500 mm. below normal (Fig. 5, Plate 1), the growth became appreciably decreased. At normal pressure the growth regained its former value.

The seedling of *Helianthus* is comparatively thicker and hardier than that of *Cajanus* and its normal rate of growth is also comparatively slower. This may possibly be a reason for its greater resistance to reduced pressure. The result of our next experiment on the lower internode of *Cajanus* will also support this deduction. The growth of the lower internode is very slow in comparison with the upper internode. The growth of such internodes was found to remain unaffected at the pressure of 360 mm. or 400 mm. below normal and was only slightly decreased at 260 mm. or 500 mm. below normal. A typical record of the decrease of growth of the lower internode of *Cajanus* in reduced pressure is given in figure 6 (Plate 1).

At 260 mm. or 500 mm. below normal (Fig. 6, Plate 1), the growth became slightly decreased. At normal pressure there was recovery.

Investigation was next undertaken to determine how the growth is affected by reduced pressure when the partial pressure of oxygen is maintained equal to that what it is in the normal atmosphere. In doing so a few experiments were performed in 260 mm. or 500 mm. below normal, the partial pressure of oxygen being 152 mm.

The growth rates of both *Cajanus* and *Helianthus* were found to remain unchanged at the reduced pressure of 260 mm. or 500 mm. below normal when the partial pressure of oxygen was maintained equal as that in the normal atmosphere. Typical records of the growth of *Cajanus* and *Helianthus* under above condition are given in figures 7 and 8 (Plate 2), respectively. In regard to the effect of purely diminished external pressure reference to figures 4 and 5 (Plate 1) will show that a pressure of 260 mm. or 500 mm. below normal causes an immediate inhibition of the rate of growth of both *Cajanus* and *Helianthus*. It is therefore clear from figures 7 and 8 (Plate 2) that the effect of reduced external pressure is only due to the diminution of the partial pressure of oxygen.

Effect of Reduced Pressure on the Growth of Roots.

It will be evident from the results of our experiments on the effect of reduced pressure on the shoots of plants that the minimum effective reduced pressure in inhibiting growth is not exactly the same in different segements of the same shoot. We, therefore, took up the investigation on the roots of plants which is structurally and functionally so different from the shoot, so as to find out how its growth is affected by reduced pressure.

For studying the effect of reduced pressure on the growth of the root, the primary root of *Cicer æritinum* and the underground root of *Pandanus odoratissimus* were employed. In employing the primary root of *Cicer* for the experiment it was allowed to grow to about a centimeter after the germination of the *Cicer* seed.

Method of Experiments.

The device which was employed to record the growth elongation of the root was exactly the same as employed in recording the embryonic growth of the seed.[5] The complete apparatus, consisted of the single Lever Recorder as employed in recording the growth of the stem, and in addition a seed holder or a clamp and a spoon shaped aluminium pan for the root tip to press upon. The seed-holder was employed in the experiments on the primary root of *Cicer*. In the experiments on the root of *Pandanus* a clamp was used in its stead for holding the root. The seed-holder is a rigid metallic piece having three silver pins against which the flat end of the seed is attached. The projected primary root presses against the spoon shaped pan which is prolonged and serves as a lever being supported by means of a fork provided with jewel bearings. The pan is attached by a thread to the recording lever so that a total magnification of one-hundred times is thus produced. A diagrammatic representation of the device is given in figure 2.

The growth elongation of the root exerts a pressure on the pan which pulls down the recording lever. The growth elongation of the root is therefore, represented by the down curve in the records.

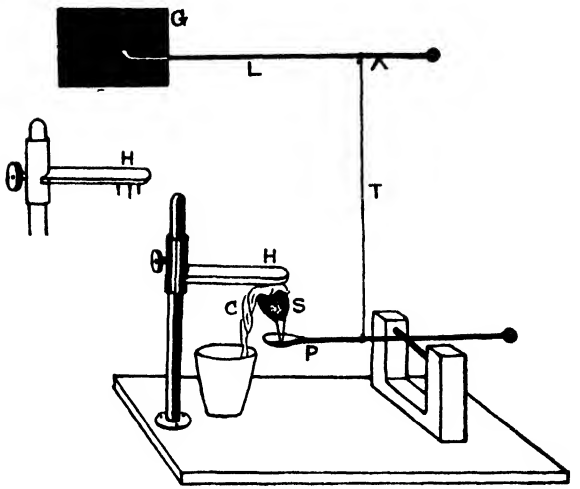


FIG. 2. Diagrammatic representation of the recording device of the growth of the primary root of *Cicer*.

C, moist cotton strip wrapped at the basal part of the seed S, which is attached to the holder H. P, the aluminium pan on which is supported the tip of the primary root of the seed. The pan is attached by a thread T, to the lever of the recorder L. The growth of the primary root is recorded on the smoked glass plate G.

The root of *Pandanus* after being carefully extricated out of the soil was kept in a dark moist chamber for two hours in order to ensure full recovery from the stimulus of mechanical disturbances after which the experiment was commenced. The germinated seed of *Cicer* with its primary root was also treated in the same manner before starting the experiment.

In observing the effect of reduced pressure on the growth of root its normal growth rate was first recorded in normal pressure previous to the pressure being reduced to a certain definite value, similarly as was done in the experiments on the shoot of the plant.

In the primary root of *Cicer* the growth appreciably decreased at a pressure of 460 mm. or at 300 mm. below normal and at 360 mm. or 400 mm. below normal the root ceased to grow. Typical records of the variation of the growth of the primary roots of *Cicer* in reduced pressure are given in figures 9 and 10 (Plate 2). Reduction of pressure is marked with an arrow and the restoration to normal pressure with an arrow

within a circle. The description of the experiments are given below.

At 460 mm. or 300 mm. below normal the rate of growth is found to be diminished after 5 minutes of the reduction of the pressure. After restoration to normal pressure the growth is found to have fully recovered.

At 360 mm. or 400 mm. below normal the growth of the root is ceased and is represented by the horizontal line in the record. After restoration to normal pressure, however, the growth is found to have almost fully recovered.

In the root of *Pandanus* the growth is found to have remained unaffected at 460 mm. or 300 mm. below normal and is only partially diminished at 360 mm. or 400 mm. below normal. Even at 260 mm. or 500 mm. below normal, the activity of growth is not entirely abolished. Whereas, reference to figure 10 (Plate 2) will show that the growth of the primary root of *Cicer* ceased at 360 mm. or 400 mm. below normal. The diminution of growth of the root of *Pandanus* is evidenced in figures 11 and 12 (Plate 3). The description of the experiments are given below.

At 360 mm. or 400 mm. below normal the growth is found to have slightly diminished as is indicated by the slight change of inclination of the curve (Fig. 11, Plate 3). After the restoration of the normal pressure the growth regained its former value. At 260 mm. or 500 mm. below normal the growth is found to have almost stopped (Fig. 12, Plate 3). Slight inclination of the curve in reduced pressure shows that the growth is not entirely stopped. After restoration of the normal pressure there was complete recovery.

In order to investigate how the growth of root is affected by reduced pressure when the partial pressure of oxygen is maintained equal to that in normal atmosphere, a few experiments were undertaken with both the primary root of *Cicer* and the underground root of *Pandanus* in as low a pressure as 260 mm. or 500 mm. below normal, partial pressure of oxygen remaining equal to that in the normal atmosphere. The normal growth of the root was found to remain absolutely unchanged under the conditions. Typical records are given in figures 13 and 14 (Plate 3).

Reference to figures 10 and 12 (Plate 3) will show how the growths of the roots of *Cicer* and *Pandanus* are diminished at 360 mm. and 260 mm. respectively. It will be evident by comparing them with figures 13 and 14 (Plate 3) that diminution of the growth of the roots in reduced pressure is absolutely due to the reduction of the partial pressure of oxygen.

From comparison of the results of the effect of reduced pressure on the growth of roots it will be evident that the primary root of *Cicer* is more susceptible to reduced pressure in comparison with that of *Pandanus*. The possible explanation is that the underground root of *Pandanus* is comparatively slow growing and hardier than the primary root of *Cicer*.

Summary.

Investigation was undertaken to determine how the growth of plant is affected by the reduction of atmospheric pressure. Observations on the effect of reduced pressures were separately recorded on the growth of both shoots and roots of different plants.

In observing the effect on shoot changes on the growth of young and old regions of the shoots of similar plants were separately recorded.

The growth of the young seedling of *Cajanus* is appreciably diminished at a pressure of 360 mm. or 400 mm. below normal and at 260 mm. or 500 mm. below normal the growth is absolutely annulled. But the growth of seedling of *Helianthus* remains unaffected up to 360 mm. or 400 mm. below normal and is only appreciably decreased at 260 mm. or 500 mm. below normal.

The growth of the lower internode of *Cajanus* remains absolutely unaffected at 360 mm. or 400 mm. below normal and is only slightly affected at 260 mm. or 500 mm. below normal.

When the partial pressure of oxygen is maintained equal to that in the normal atmosphere the growth of the seedlings of both *Cajanus* and *Helianthus* remains unaffected even though the total pressure was reduced to 260 mm. or 500 mm. below normal.

The growth of the primary root of *Cicer* is appreciably decreased at 460 mm. or 300 mm. below normal and at 360 mm. or 400 mm. below normal the growth is absolutely annulled.

The growth of the underground root of *Pandanus* is only slightly diminished at 360 mm. or 400 mm. below normal and even at 260 mm. or 500 mm. below normal the growth is not absolutely stopped.

The growth of the roots of both *Cicer* and of *Pandanus* remains unaffected even in a reduced pressure of 260 mm. or 500 mm. below normal when the partial pressure of oxygen in it is maintained equal as that in the normal atmosphere.

REFERENCES.

[1] B. K. Dutt and A. Guha-Thakurta.—Investigation on Physiological Reactions of Plants under Low Atmospheric Pressure, Trans. Bose Res. Inst., Vol. XI, pp. 55–69, 1934-35.

[2] Charles J. Whitfield.—Ecological Aspect of Transpiration, Pikes peak region: climatic aspects, Bot. Gaz. 93, pp. 437–452, 1932.

[3] Shope Paul F.—Stem and Leaf Structure of Aspen at different Altitudes in Colorado, Amer. Jour. Bot. 14(3), pp. 116–119, 1927.

[4] P. W. Wilson.—Effect of Pressure of Atmosphere on Development of Red Clover, Nature, Vol. 136, pp. 262-63, 1935.

[5] B. K. Dutt and A. Guha-Thakurta.—Automatic Record of Embryonic Growth on Germination of seed, Trans. Bose Res. Inst., Vol. IV, pp. 58–76.

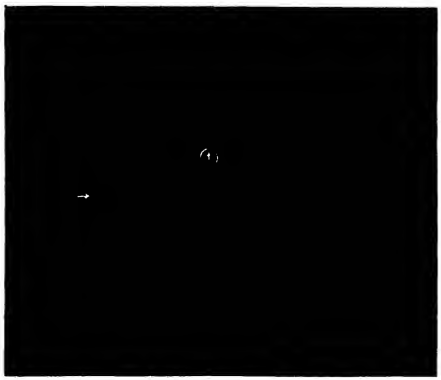


FIG. 3.

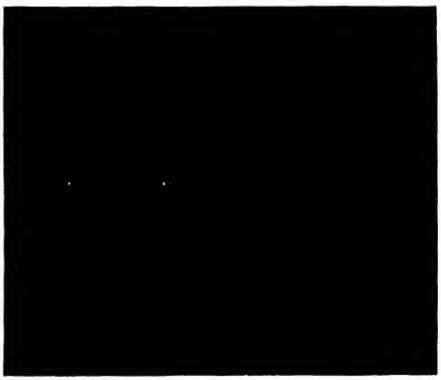


FIG. 4.

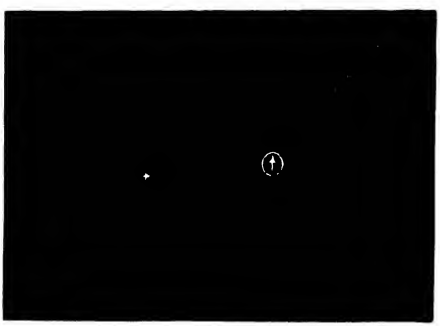


FIG. 5.

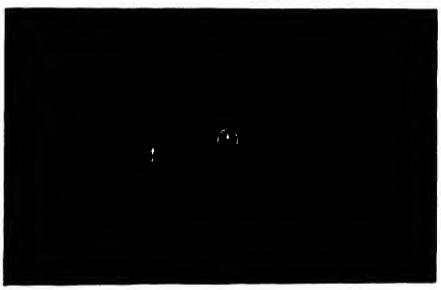


FIG. 6.

FIG. 3. Effect of reduced pressure of 360 mm. or 400 mm. below normal on the growth of the shoot of *Cajanus*.
The pressure was reduced at arrow and the reduced pressure withdrawn at arrow within circle. The dots are at intervals of 30 seconds.

FIG. 4. Effect of reduced pressure of 260 mm. or 500 mm. below normal on the growth of the shoot of *Cajanus*.

FIG. 5. Effect of reduced pressure of 260 mm. or 500 mm. below normal on the growth of the shoot of *Helianthus*.

FIG. 6. Effect of reduced pressure of 260 mm. or 500 mm. below normal on the growth of the lower internode of *Cajanus*.

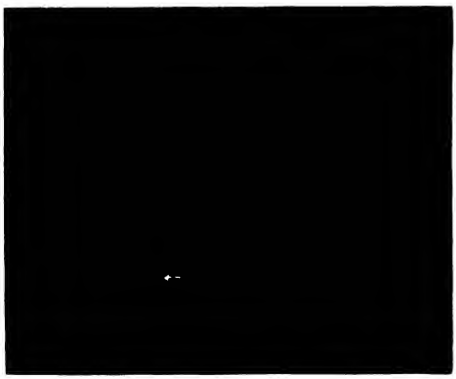


FIG. 7.

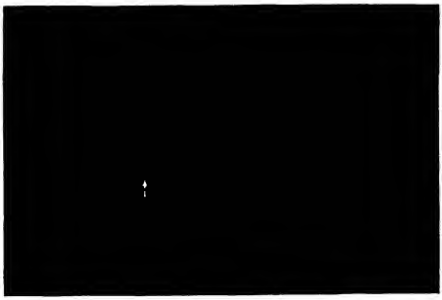


FIG. 8.

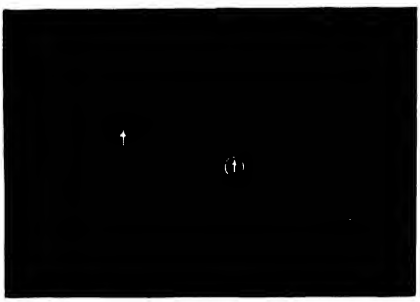


FIG. 9.

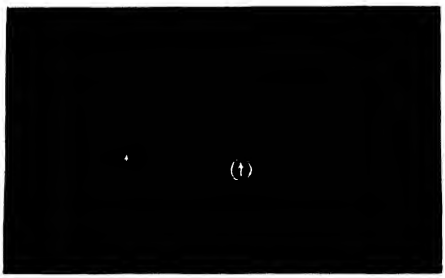


FIG. 10.

FIG. 7. Effect of reduced pressure of 260 mm. or 500 mm. below normal on the growth of the shoot of *Cajanus* when the partial pressure of oxygen is maintained equal to that of the normal atmosphere.
Note that the normal rate of growth remains unchanged in the reduced pressure.

FIG. 8. Effect of reduced pressure of 260 mm. or 500 mm. below normal on the growth of *Helianthus* when the partial pressure of oxygen is maintained equal to that of the normal atmosphere.
Note that the normal rate of growth remains unchanged in the reduced pressure.

FIG. 9. Effect of reduced pressure of 460 mm. or 300 mm. below normal on the growth of the primary root of *Cicer*.
The pressure was reduced at arrow and the reduced pressure was withdrawn at arrow within circle.

FIG. 10. Effect of reduced pressure of 360 mm. or 400 mm. below normal on the growth of the primary root of *Cicer*.

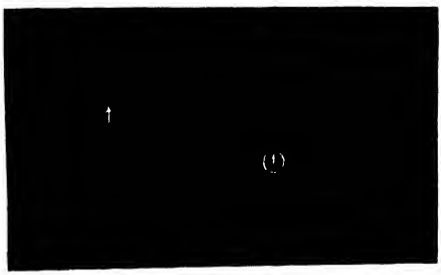


FIG. 11.

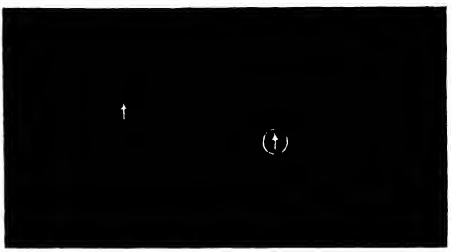


FIG. 12.

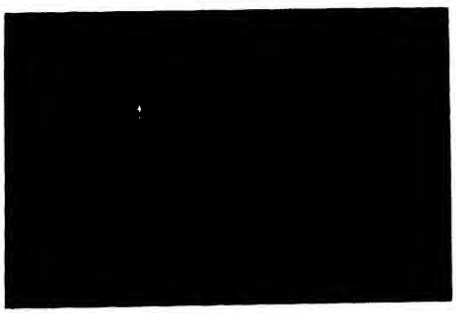


FIG. 13.

FIG. 14.

FIG. 11. Effect of reduced pressure of 360 mm. or 400 mm. below normal on the growth of the underground root of *Pandanus*.

FIG. 12. Effect of reduced pressure of 260 mm. or 500 mm. below normal on the growth of the underground root of *Pandanus*.

FIG. 13. Effect of reduced pressure of 260 mm. or 500 mm. below normal on the growth of the primary root of *Cicer* when the partial pressure of oxygen is maintained equal to that of the normal atmosphere.

Note that the normal rate of growth remains unchanged in the reduced pressure.

FIG. 14. Effect of reduced pressure of 260 mm. or 500 mm. below normal on the growth of the underground root of *Pandanus* when the partial pressure of oxygen is maintained equal to that of the normal atmosphere.

Note that the normal rate of growth remains unchanged in the reduced pressure.

II. INVESTIGATION ON THE INFLUENCE OF LIGHT AND TEMPERATURE ON THE DIURNAL VARIATION OF GROWTH.

By A. GUHA-THAKURTA and B. K. DUTT.

(*Received for publication 15th August, 1938.*)

In his investigation on the diurnal movements of various anisotropic plant organs Sir J. C. Bose [1] has shown that amongst the numerous factors which complicate the diurnal movements of plants by the algebraic summation of their effects the most important are the effects of light and darkness, of variation of temperature and of thermal variation on organs subjected to the action of gravity. He has also shown by his method of short period observations that the growth of plant also is profoundly affected by light and darkness and variation of temperature. Regarding the effects of light and temperature on growth he has further come to the conclusion that their effects are antagonistic to each other; light normally brings about a decrease of the rate of growth whereas increase of temperature up to a certain limit accelerates the rate of growth.

The object of our present investigation is to determine the exact nature of variation of the diurnal growth of plant and how that variation is related to the individual effect of light and of temperature.

Different workers have observed a periodic variation in the rate of diurnal growth of plant. Omaston [2] noted that the increase in height of bamboo was 9·65 ft. in 14 days of which 3·7 ft. occurred by day and 5·95 ft. by night. Brown and Trelease [3] found that the shoots of *Cestrum nocturnum* which is one of the most rapidly growing plants around Manila, P.I., decreased in length during the day but that later in the afternoon they returned to their original length and increased during the night. Prescott [4] found that the growth rate of corn in Egypt as measured by the increase in height was generally greater during the 12 hours of night from 8 P.M. to 8 A.M. than that during the

remaining 12 hours of the day. Miller [5] observed at Manhattan, Kans., that corn plants which were about 4 ft. high increased in height 6 cm. from 7 A.M. to 7 P.M. and 8 cm. from 7 P.M. to 7 A.M. Mason [6] reported that in the date palm normal growth, which is manifest by the pushing up of the leaves from the growth centre, is made chiefly in the time between sunset and sunrise.

Poterfield [7] reported that the ratio of the night growth of bamboo to that of daylight was 1·8 to 2·8 as reported by investigators in Java, Ceylon and India. In Algiers, however, the growth of this plant was greater during the day than during the night. Poterfield found the ratio of the day growth to night growth to be 1·6.

It appears from the above reports that all of the workers have observed an enhanced rate of growth during the night with the only exception of Poterfield. From his report it appears that the growth of bamboo is enhanced during the day in Algiers instead of during the night at Java, Ceylon and India. It seems curious that the same plant exhibits entirely different habits at different places.

Conclusions arrived at by Sir J. C. Bose on the individual effects of light and temperature on growth may help to some extent to explain the periodic variation of growth. The greater increase during the night and less during the day can be attributed to the overpowering influence of absence of light on the inhibiting effect of lower temperature. Cases of enhanced growth during day may be explained by higher temperature effect overpowering the inhibitory effect of light.

Careful analysis of the diurnal periodicity of growth may help to some extent to bring out the predominant factor. But the absolute effects of the factors can only be determined from the isolation of the individual effects from the resultant effects of the factors. The isolation of the absolute effects of any one of the factors can be possible by keeping one of the factors constant during twenty-four hours of the day and recording the effect due to diurnal variation of the other factor.

The intensity and duration of these two external factors does not remain constant throughout the year but changes at different

seasons. In determining the individual influence of the factors on the diurnal variation of growth it is, therefore, necessary to investigate the individual influence of the factors at different seasons.

In investigating the influences of light and temperature on the diurnal growth of plant the subjects to be treated in this paper are the following:—

(I) Investigation on the influence of light and temperature on the diurnal growth of plants in summer.

(II) Investigation on the diurnal growth in winter.

The objects of the investigations in each of the above subjects necessitated the following order of experiments:—

1. Recording of the diurnal growth under normal variation of light and temperature.
2. Recording of the diurnal growth under constant light and temperature.
3. Determination of the diurnal growth in constant temperature under normal variation of light.
4. Determination of the diurnal growth in constant intensity of light under normal variation of temperature.

Method of Experiment.

Seedlings of *Cajanus* were employed for experimental purposes. The seeds after germination were transferred in pots containing earth. For each of the above series of experiments seedlings of a definite age were employed. A single specimen was not repeatedly used to complete a series of experiments, because the tonic condition of the specimen which changes with age might introduce complications to study the absolute effects of the factors under investigation. Each specimen was used for observation of the variation of growth for twenty-four hours only; therefore a number of specimens were required to complete a series of experiments. The selected specimens for each series of experiments were not only of the same age but of similar height and vigour.

The pot containing the plant was supplied with sufficient quantity of water so that the suctional activity suffered no

change. A big tray containing water was also kept in the vicinity of the plant so as to maintain more or less a uniform condition of humidity around the plant. A Recording Hygrometer kept near the plant showed no appreciable diurnal variation of the hygrometric state of the atmosphere, under the condition.

Observations were made by directly recording the diurnal variation of the growth of the plant for twenty-four hours by a Single Lever Oscillating Plate Recorder. The plant was attached to the recording lever of the recorder by a fine silk thread. The growth elongation of the stem was indicated by the up-movement of the lever. The growth of the plant was automatically recorded by a series of dots on the smoked glass plate of the recorder. The dots were recorded at intervals of fifteen minutes. The diurnal record of growth, therefore, consisted of 96 dots in twenty-four hours.

The daily fluctuation of temperature was recorded by a maximum-minimum thermometer.

For maintenance of constant temperature an electro-thermostatic glass chamber constructed in the Institute was employed. The chamber was maintained at a constant temperature by a thermo-electric regulator which interrupts the heating current as soon as the temperature of the chamber is raised just above the predetermined constant temperature. The make and break of the current takes place automatically so that the temperature of the chamber is maintained constant throughout day and night. In maintaining the temperature constant the maximum temperature of the season was taken into account. The chamber being made of glass the entrance of light was not obstructed in any way. The plant and the recording apparatus were placed inside the closed chamber when recording in constant temperature was necessary.

In maintaining constant light for twenty-four hours a 500 c.p. electric lamp was employed. The light was allowed to fall vertically on the plant from above. The plant was enclosed in an open-mouthed metallic cylinder to enable it to receive light from the artificial source alone. The lamp was kept at such a distance from the plant so that the intensity of the constant light was

equal to that of the mid-day diffused light of the room. The intensity was determined by a photo-electric cell.

Influence of Light and Temperature on the Diurnal Growth in Summer.

Observations in summer were recorded between 2nd August and 12th September. The maximum room temperature during the period did not exceed 88°F, while the minimum temperature at night did not fall below 77°F. The total diurnal fluctuation of temperature during the period was 8°F to 10°F. The period of day was considerably longer than that of night. The time of sunrise shifted from 5-33 A.M. on 2nd of August to 5-47 A.M. on 12th of September and that of sunset from 6-39 P.M. to 6-5 P.M.

The seedlings, 6 days old from the time of germination, were employed for this series of experiments. Such seedlings attained a height of about 6 cm. within that period.

In arriving at a definite conclusion regarding the individual effects of light and temperature on the diurnal growth the experiments were undertaken in the order previously described. The series of experiments were repeated several times over for full confirmation of the results. Each kind of experiment has been represented by a typical record. The normal growth of the seedlings being quite appreciable a magnification of only two was employed for recording.

Recording of the Diurnal Growth under Normal Variation of Light and Temperature.

Experiment 1. Object of this experiment is to determine the exact nature of the diurnal growth in summer. The diurnal record (Fig. 1, Plate 4) shows that the growth during the night was much enhanced in comparison with that during the day. The enhancement of growth commenced from 6-30 P.M. and enhanced rate persisted up to 6-30 A.M. After that the rate was gradually decreased. The rate of growth between 9 to 12 A.M. is more or less equal to that of previous day's growth between 12 A.M. to 5 P.M. recorded in the first part of the curve. The

maximum and minimum temperatures during 24 hours were 86·5°F and 77°F respectively, the total fluctuation being 8·5°F.

Recording of the Diurnal Growth under Constant Temperature and Light.

Experiment 2. This experiment will show whether light and temperature are the only important factors responsible for the periodicity of diurnal growth. If so, the periodicity will be absent in the diurnal growth and the plant will continue to grow in an uniform rate when these factors are maintained constant. But in presence of any other important factors which influence the periodicity, the growth rate will not be uniform in spite of the light and temperature being constant.

It will be evident from the diurnal growth curve (Fig. 2, Plate 4) under constant light and temperature that the rate of growth has remained more or less unchanged throughout twenty-four hours. This indicates that light and temperature are the only important factors which influence the periodic variation of diurnal growth.

This being conclusively proved from the above experiment that no other external factor besides light and temperature contributes in a substantial way to cause the periodicity of diurnal growth we next took up the investigation to determine how light and temperature individually influence the diurnal growth by two subsequent experiments: One by recording the diurnal growth in constant temperature under normal variation of light, and the other in constant light under normal variation of temperature.

Determination of the Diurnal Growth in Constant Temperature under Normal Variation of Light.

Experiment 3. In this experiment the temperature was kept constant and the normal variation of light was the only factor that remained to influence the diurnal growth. The diurnal variation of growth under such condition can, therefore, be taken as the absolute effect due to light and absence of light.

Temperature was maintained constant at 88°F which was 1°F higher than the maximum temperature during the period when this experiment was conducted.

The record of the diurnal variation of growth (Fig. 3, Plate 4) shows that the growth in the absence of light was much greater than that during the day. The enhanced rate commenced from 7 P.M. and persisted up to 6-30 A.M. Reference to figure 1 shows also that the growth rate enhanced during the night, but the enhancement under that condition was comparatively less than that obtained in figure 3 (Plate 4). The greater enhancement of growth in figure 3 (Plate 4) can be reasonably attributed to the absence of inhibiting influence of lower temperature of night.

Determination of Diurnal Growth in Constant Light under Normal Variation of Temperature.

Experiment 4. In this experiment, light being kept constant, the diurnal growth was influenced by variation of temperature alone. The maximum temperature of the day was 88°F and the minimum temperature at night was 78·5°F, the total fluctuation being 9·5°F.

The record (Fig. 4, Plate 5) shows that the rate of growth during the night was comparatively less than that during the day. The decrease of growth begins from 6-30 P.M. and persists up to 6-30 A.M. From 6-30 A.M. the rate of growth is gradually increased.

From comparison of the individual influence of light and temperature represented by the figures 3 (Plate 4) and 4 (Plate 5), respectively, it will be clear that the effect of light is more important than that of the temperature in bringing about the variation of diurnal growth. The growth of the plant is inhibited during the day and accelerated during the night due to the presence and absence of light alone, while due to the individual influence of temperature growth is slightly accelerated during the day, and inhibited during the night. That the effect of light overpowers the effect of temperature will be clear from figure 1 which represents the diurnal growth under normal variation of light and temperature.

The actual diurnal growth and the average rates of growth during both day and night as obtained in different experiments in summer are given in Table I. For convenience of inspection, results of similar experiments are grouped together in the table; each group containing results of five experiments.

TABLE I.

Diurnal growth of Cajanus seedlings under different conditions of light and temperature variation in summer.

Condition.	Variation of temperature during 24 hours in °F.		Total growth in 24 hours.	Average day growth per hour.	Average night growth per hour.	Ratio of night growth to day growth.
	Max.	Min.				
Under normal variation of light and temperature.	86°F	78°F	48 mm.	1·5 mm.	2·5 mm.	1·66
	88°F	78°F	47 ,,	1·45 ,,	2·42 ,,	1·67
	87·5°F	77·5°F	46 ,,	1·42 ,,	2·39 ,,	1·68
	86·5°F	77°F	47 ,,	1·51 ,,	2·5 ,,	1·65
	86°F	77°F	47·5 ,,	1·5 ,,	2·5 ,,	1·66
Under constant temperature and light.	Temperature constant at 88°F.		40 ,,	1·66 ,,	1·66 ,,	1
			42·5 ,,	1·77 ,,	1·77 ,,	1
			43 ,,	1·79 ,,	1·79 ,,	1
			41 ,,	1·5 ,,	1·5 ,,	1
			44 ,,	1·8 ,,	1·8 ,,	1
Under constant temperature and normal variation of light.	Temperature constant at 88°F.		52·5 ,,	1·51 ,,	2·9 ,,	1·91
			52 ,,	1·42 ,,	3 ,,	2·11
			50 ,,	1·48 ,,	2·7 ,,	1·82
			52·5 ,,	1·5 ,,	2·9 ,,	1·93
			53 ,,	1·6 ,,	2·87 ,,	1·79
Under constant intensity of light and normal variation of temperature.	87°F	78°F	38 ,,	1·75 ,,	1·45 ,,	0·82
	87·5°F	78°F	37 ,,	1·71 ,,	1·38 ,,	0·80
	86°F	77°F	33 ,,	1·61 ,,	1·14 ,,	0·70
	88°F	78·5°F	36 ,,	1·62 ,,	1·25 ,,	0·70
	86·5°F	77°F	34 ,,	1·62 ,,	1·25 ,,	0·70

In the above table the total average growth under normal variation of light and temperature is 47·1 mm.; under constant light and temperature 42·1 mm., under constant temperature and normal variation of light 52 mm., under constant light and normal variation of temperature 35·6 mm.

It will, therefore, be evident that during the summer the maximum diurnal growth was obtained under conditions of constant temperature and normal variation of light; the minimum

diurnal growth was obtained under conditions of constant light and normal variation of temperature. Under constant temperature and constant light the total diurnal growth was less than that obtained under normal variation of these two factors.

Investigation on the Influence of Light and Temperature on Diurnal Growth in Winter.

The experiments in winter were completed between the 22nd November and the 22nd December, 1937. The time of sunrise shifted from 6-21 on the 22nd November to 6-41 on the 22nd December and that of sunset from 5-10 to 5-14 correspondingly. The maximum temperature, as recorded, in the experimental room rose up to 77°F and the minimum fell to 60°F during the period. The total fluctuation of the diurnal temperature varied from 12° to 15°F.

The time of germination of the seed was longer in winter than in summer. The growth of the seedlings being comparatively slower in this season, they were selected for experiment when 9 days old after germination; after this period they were almost equal in height to the 6 days old summer specimens.

The experiments were conducted in the same order as adopted in summer. Typical experiments are described below.

Recording of the Diurnal Growth under Normal Variation of Light and Temperature in Winter.

Experiment 5. The maximum temperature of the room rose up to 75°F. and minimum fell to 63°F at night, the total fluctuation of temperature being thus 12°F.

The diurnal record (Fig. 5, Plate 5) shows that the growth during the night was comparatively less than that during the day. The decrease of rate of growth during the night in winter indicates that the effect of low temperature overpowered the effect of absence of light.

The resultant effects of light and temperature on diurnal growth of the plant are, therefore, different in different seasons. In summer when the temperature fluctuates at higher range of the thermometer scale, the effect of temperature is overpowered by the effect of light and absence of light, and in winter when

the temperature fluctuates at lower range of the thermometer scale the effect of temperature overpowers the effect of light and its absence.

Record of the Diurnal Growth under Constant Temperature and Light in Winter.

Experiment 6. Temperature was kept constant at 77°F which was 1°F above the maximum temperature of the room. The intensity of the constant light was the same as that of the midday diffused light of the room; this was found to be almost equal to the intensity in summer.

The record (Fig. 6, Plate 5) shows that the growth continued in an uniform rate during both day and night. This indicates that there was no other factor present to induce variation of diurnal growth besides light and temperature.

Determination of the Diurnal Growth at Constant Temperature under Normal Variation of Light in Winter.

Experiment 7. From this experiment the absolute effect of variation of light in inducing any change in diurnal growth in winter will be evidenced. The temperature was kept constant at 77°F which was 1·5°F above the maximum temperature of the day and the diurnal growth was recorded under normal variation of light alone.

The record (Fig. 7, Plate 6) shows that the rate of growth increased during the night in the absence of light. From comparison of the record with figure 5 which represents the diurnal growth in winter under normal variation of light and temperature, it will be clear how the accelerating influence of the absence of light has been overpowered by the inhibiting influence of low temperature during the night in winter.

Determination of the Diurnal Growth in Constant Light under Normal Variation of Temperature in Winter.

Experiment 8. From this experiment the absolute effect of temperature in inducing a change in diurnal growth in winter will be evidenced. The intensity of light being constant during

both day and night the changes in the diurnal growth will be due to the variation of temperature alone. The maximum temperature of the day was 76°F and the minimum temperature at night 63°F, the total fluctuation being 13°F. The record (Fig. 8, Plate 6) shows that the rate of growth has greatly decreased during the night. From a comparison of this record with that of figure 5, Plate 5, which represents the diurnal growth under normal variation of light and temperature in winter, it will be seen that the diminution of the rate of growth at low temperature of night has been more pronounced when freed from the accelerating influence of absence of light.

The actual diurnal growth and the average rates of day and night growths as obtained in different experiments in winter are given in Table II.

TABLE II.

Diurnal Growth of Cajanus seedlings under different conditions of light and temperature variation in winter.

Condition.	Variation of temperature during 24 hrs. in °F.		Total growth in 24 hours.	Average day growth per hour.	Average night growth per hour.	Ratio of night growth to day growth.
	Max.	Min.				
Under normal variation of light and temperature.	75°F	63°F	26·5 mm.	1·29 mm.	0·83 mm.	0·64
	76·5°F	64°F	25 ,,	1·2 ,,	0·89 ,,	0·74
	75·5°F	62°F	26 ,,	1·25 ,,	0·92 ,,	0·73
	76°F	62°F	23·5 ,,	1·12 ,,	0·83 ,,	0·74
	71·5°F	61·5°F	25 ,,	1·16 ,,	0·91 ,,	0·78
Under constant temperature and light.	Temperature constant at 77°F.		31 ,,	1·29 ,,	1·29 ,,	1
			29·5 ,,	1·22 ,,	1·22 ,,	1
			31·5 ,,	1·31 ,,	1·31 ,,	1
			31 ,,	1·29 ,,	1·29 ,,	1
			28 ,,	1·16 ,,	1·16 ,,	1
Under constant temperature and normal variation of light.	Temperature constant at 77°F.		35 ,,	1·15 ,,	1·78 ,,	1·46
			35·5 ,,	1·16 ,,	1·80 ,,	1·55
			36·5 ,,	1·3 ,,	1·75 ,,	1·34
			36·5 ,,	1·2 ,,	1·91 ,,	1·59
			33·5 ,,	1·2 ,,	1·75 ,,	1·45
Under constant intensity of light and normal variation of temperature.	76°F	63°F	17·5 ,,	1·02 ,,	0·45 ,,	0·44
	76°F	62·5°F	18·5 ,,	1·0 ,,	0·54 ,,	0·54
	76·5°F	63·F	18·0 ,,	0·9 ,,	0·54 ,,	0·60
	74°F	60°F	18·5 ,,	0·9 ,,	0·58 ,,	0·64
	74·5°F	62°F	18·5 ,,	1·04 ,,	0·54 ,,	0·51

In the above table the total average growth under normal variation of light and temperature is 25·2 mm., under constant temperature and light it is 30·2 mm., under constant temperature and normal variation of light it is 35·4 mm. and under constant light and normal variation of temperature it is 18·2 mm.

In analysing the results contained in Tables I and II it is to be found that the average ratio of night growth to day growth in winter is 0·74 as against 1·66 in summer, under normal variation of light and temperature. Under constant light and temperature, though the rate of growth is different in summer and winter, the ratio of night growth to day growth is 1 in both the seasons. Under constant temperature and normal variation of light, growth is increased during the night in both the seasons, but the ratio of night growth to day growth is comparatively higher in summer than in winter; the higher ratio in summer may be reasonably attributed to the higher value of the temperature constant of the season. Under constant light and normal variation of temperature the growth is decreased during the night in both the seasons but the ratio of night growth to day growth is comparatively lower in winter than in summer; the lower ratio in winter may be attributed to the influence of lower temperature.

Summary.

The principal object of the present investigation is to discriminate the individual influences of light and of temperature from the summated effects of both, in inducing the diurnal variation of growth.

Diurnal record of growth under normal variation of light and temperature in summer shows that there was increase of growth during the night, indicating that the accelerating influence of the absence of light overpowered the inhibiting influence of the lower temperature of night.

Under constant light and temperature the growth continued in an uniformly equal rate during both day and night indicating that there was no other factor present besides light and temperature to induce variation of the rate of growth.

Under constant temperature and normal variation of light the increase of growth during the night was greater in com-

parison with that under normal variation of both of these factors; the greater increase during the night was due to the absence of the inhibiting influence of lower temperature of night.

Under constant light and normal variation of temperature the rate of growth is slightly decreased during the night; slight decrease was only due to the inhibiting influence of lower temperature during the night under the condition.

Diurnal record of growth under normal variation of light and temperature in winter shows that there was decrease of growth during the night, indicating that the inhibiting influence of low temperature of winter night overpowered the accelerating influence of absence of light.

Under constant light and temperature in winter the rate of growth was uniform during both day and night.

Under constant temperature and normal variation of light the growth greatly increased during the night in winter; the inhibiting influence of falling temperature being absent the increase of growth was only due to the influence of absence of light.

When the light was kept constant the growth greatly decreased during the night under normal variation of temperature in winter; the accelerating influence of the absence of light being withdrawn at night, the decrease of growth was wholly due to the inhibiting influence of lower temperature of winter night.

REFERENCES.

[1] Bose, Sir J. C.—Growth and Tropic Movements of Plants. 1929.

[2] Omaston, B. B., 1918—Rate of Growth of Bamboos, Indian Forester, 44, pp. 52–57; Exp. Sta. Rec., 44, pp. 518-519.

[3] Brown, W. H., and Trelease, S. F., 1918—Alternate Shrinkage and Elongation of Growing Stems of *Cestrum nocturnum*, Phil. Jour. Sci., C. 13, pp. 353–360.

[4] Prescott, J. A., 1921.—Some Observations on the Growth of Maize in Egypt, Sultanic Agr. Soc. Bull., 7.

[5] Miller, E. C., 1925.—Unpub. data, Kans. Agr. Exp. Sta.

[6] Mason, S. C., 1925.—Partial Thermonasty of the Growth Centre of the Date Palm, Jour. Agr. Res., 31, pp. 415–455. The Inhibition Effect of Direct Sun Light on the Growth of Date Palm, Jour. Agr. Res., 31, pp. 455–469.

[7] Poterfield, W. M., 1928.—A Study of the Grand Period of Bamboo, Bull. Torrey Bot. Club, 55, pp. 327–406.

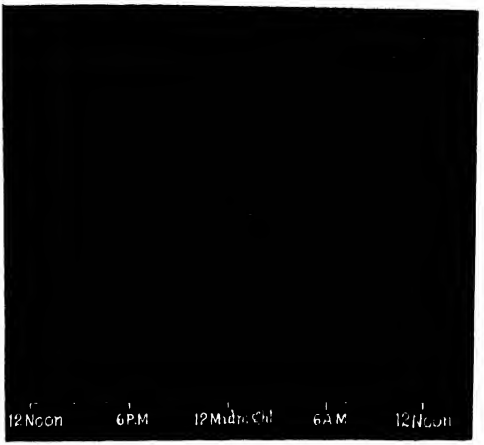


FIG. 1.

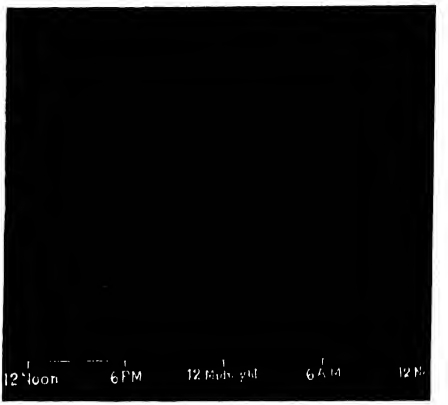


FIG. 2.

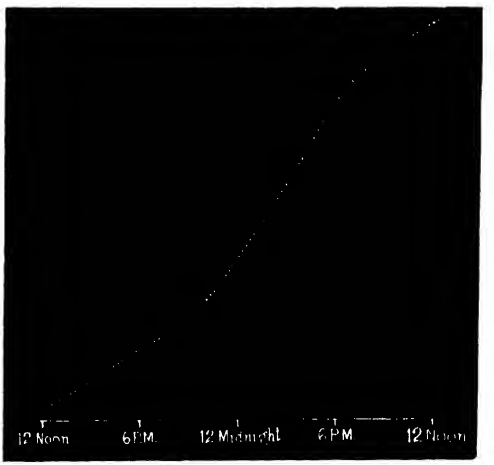


FIG. 3.

FIG. 1. Record of diurnal growth of *Cajanus* in summer under normal variation of light and temperature.

FIG. 2. Record of diurnal growth of *Cajanus* in summer under constant light and temperature.

FIG. 3. Record of diurnal growth of *Cajanus* in summer under constant temperature and normal variation of light.

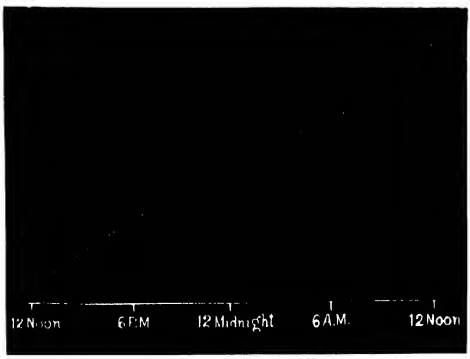


FIG. 4.

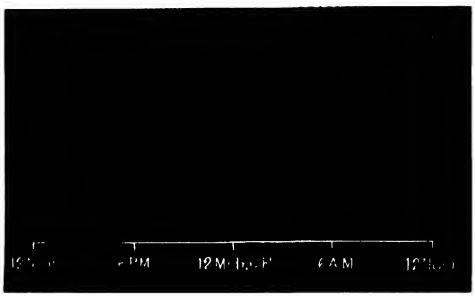


FIG. 5.

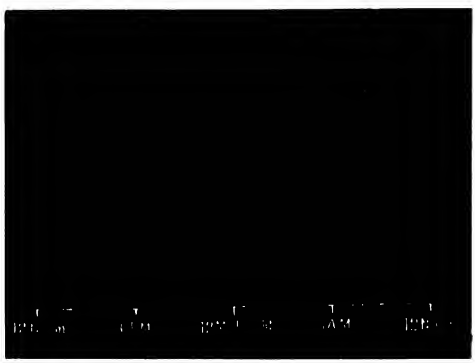


FIG. 6.

FIG. 4. Record of diurnal growth of *Cajanus* in summer under constant intensity of light and normal variation of temperature.

FIG. 5. Record of diurnal growth of *Cajanus* in winter under normal variation of light and temperature.

FIG. 6. Record of diurnal growth of *Cajanus* in winter under constant light and temperature.

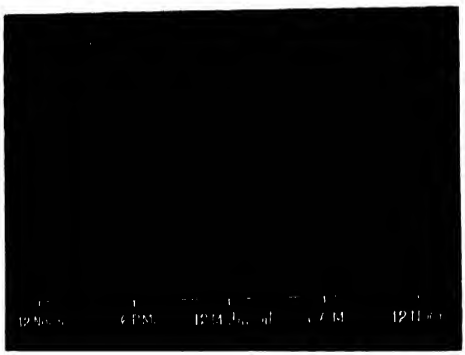


FIG. 7.

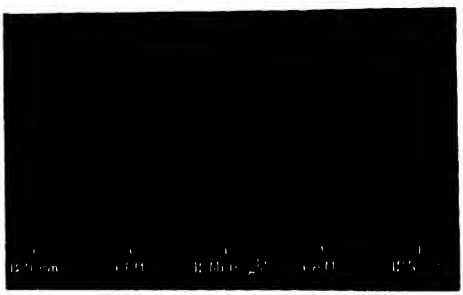


FIG. 8.

FIG. 7. Record of diurnal growth of *Cajanus* in winter under constant temperature and normal variation of light.

FIG. 8. Record of diurnal growth of *Cajanus* in winter under constant intensity of light and normal variation of temperature.

III. THE INVESTIGATION ON THE GROWTH OF LEAVES OF *VALLISNERIA SPIRALIS* WITH AND WITHOUT ROOTS.

By B. K. PALIT, B.Sc.

(*Received for publication 1st September, 1938.*)

INTRODUCTION.

The problem of studying the variation in longitudinal growth of both land and water plants in culture-solution has engaged the attention of numerous investigators. The function of roots growing in the soil consists generally in absorption, conduction, anchorage and storage. The most important amongst them is the absorption of water and inorganic salts, which is effected by the roots generally from the soil. The processes involved in the absorption of inorganic salts by the root hairs are exceedingly complex and not yet fully understood.[1] Agnes Arber[2] remarked: 'In the case of roots of water plants however, the function of anchorage has assumed a greater importance, while the function of absorption is less pre-eminent. A firm hold in the mud, and erectness of the flowering stem, are often a *sine qua non* for aquatics, and their roots help in various ways to bring this about'. Sauvageau[3] working on the transpiration current of submerged aquatics experimented upon detached branches of submerged plants, the cut end being sealed with cocoa butter and roots having been removed, he found that even under these circumstances the stem could live and develop fresh buds. Pond[4] experimenting on transpiration-current made comparative cultures of certain submerged species (Vallisneria, Elodea, etc.) rooted in soil, rooted in washed gravel, or anchored above the soil in such a way that the roots were unable to penetrate it. He found, that the rooted plants as a rule grew much better than those that were merely anchored. He further found that the retardation in growth of non-rooted plants was their inability

to secure enough phosphorus, potassium and other elements. He observed that such plants as in the case of Vallisneria, were not only stunted in growth but had their tissues loaded with an abnormal amount of starch, he came to the conclusion that lack of certain salts inhibited protein synthesis and growth though the conditions were favourable to photosynthesis. More recent work of Brown,[5] who had found that the difference in growth between rooted and unattached plants can be altogether eliminated by passing CO_2 through the water several times a day, contradicted the views of Pond. Brown considers that the non-rooted plants do not suffer at all from lack of salts but chiefly from lack of the supply of this gas which is given off from soil containing organic matter.

If it is possible during the life cycle of a plant to eliminate the soil without impairing the function of roots the problem of absorption becomes less complex. Here, however, comes the necessity of introducing nutritive culture solutions directly to the plant, because getting rid of the soil means throwing off of many nutritive substances varying according to the nature of the soil. And in using the culture-solution it must be regularly oxygenated and the balance of nutritive elements in the culture solution should be maintained, thereby retaining intact the specific effects of anions as well as of the cations.

If now the roots, whose function in the physiology of plant is so important, are removed and the plant is allowed to grow during its entire life cycle in the nutritive solution, the question arises—what will happen ? Will the plant proceed to decay or will it show better growth ? The results of the following experiments will throw a flood of light on the nature of the changes that the plant undergoes during its entire life cycle.

In our paper on 'Variation in Longitudinal growth of *Vallisneria spiralis* during its entire life cycle ',[6] it has been found that the specimen showed increase in longitudinal growth till the 9th day of the age of the leaf, the rate of growth attaining the maximum on the 9th day. After this the rate of growth began to decrease till the 15th day when growth stopped altogether.

Moreover, it has been found that in the first phase of growth-cycle the rate of growth is greater during the day time than that at night. In the next phase there was tendency towards equalisation of the rates of growth during the night and the day. Then again in the last phase the rate was found to be greater during the night than that during the daytime. Finally, the rates both at night and at day underwent diminution until stoppage of growth occurred. The solution used in the said experiment was ordinary tank water in which the plant had been acclimatised.

The object of the present inquiry is to obtain an insight into the variation in longitudinal growth of the leaf of a water plant during its entire life cycle grown in a standard nutrient solution (*a*) with roots intact, and (*b*) fibrous roots cut off. The water plant *Vallisneria spiralis* was selected for this purpose. The experiments were performed within as short a time as possible to avoid seasonal variation.

Material and Method.

The plant *Vallisneria spiralis* is a completely submerged water plant. It has a short stem. The leaves are ribbon-like and generally about a quarter inch in breadth and one and a half to two feet in length. At the base of the stem there are numerous white fibrous roots. It is reproduced mainly through stolons.

The solution used for the experiment of this series was Knop's solution for the water-culture of green plants, the composition of which is—

Calcium Nitrate	0·80 grm.
Potassium Nitrate	0·20 ,,
Potassium Dihydrogen Phosphate ..	0·20 ,,
Magnesium Sulphate	0·20 ,,
Ferric Phosphate	trace.
Distilled Water	1,000 c.c.

Experimental Arrangements.

The experiments were performed in a glass house provided with suitable arrangements for free ventilation. The temperature

of the glass house was found to undergo a diurnal variation of 8°C. Young specimens of Vallisneria of about the same age together with the mother plant were planted in earthen pots and allowed to grow in the tank of the Institute. Specimens were selected from amongst them in such a way that the central youngest leaf was of a definite length and age. In all these experiments plants with 5 other leaves and the leaf to be experimented upon were allowed to grow in special rectangular glass vessel. The stem of the specimen was fastened to the neck of a small hollow cylindrical glass flask, the inner space of which was loaded with lead shots for retaining the plant in situ at the bottom of the vessel. The upper end of the cylinder was drawn into hooks laterally on two opposite sides to prevent slipping of the specimen and then sealed. Thus tied the specimen was next placed in a beaker. The specimen with the beaker was then transferred to a rectangular glass vessel. The level of the solution in the vessel, to start with, was one and a half inches above the tip of the experimental leaf. The tip of the experimental leaf was suitably held with a clamp specially made in the form of an inverted T. The diurnal records were obtained with an Oscillating Recorder. Suitable glass links were made use of in connecting the tip of the experimental leaf with the s-shaped hook suspended by means of waxed silk thread from the writing lever of the Recorder. The plant chamber as well as the Recorder were covered with a glass case to prevent disturbance due to air current.

A semi-diagrammatic representation of the complete apparatus as well as the plant chamber is given in Fig. 1. A rectangular glass vessel R measuring 12 inches and 8 inches in length and breadth respectively and 2 feet in height was used as a plant chamber. The stem of the specimen was fastened tightly with the neck of a small cylindrical glass flask B about 2 inches in height. The plant was then introduced into a glass beaker GB whose height was 4 inches and diameter 3 in. The outer side of the beaker was covered with a black silk cloth to prevent light falling directly on the roots. The tip of the experimental leaf was held securely with a light aluminium clamp provided with two moveable pieces in the form of an inverted T. The

upper free end of T has a hole. Into this hole one end of a thin glass link was introduced and the other end of the link inserted into the s-shaped hook suspended by means of silk thread from the writing lever or of the recorder. The level of the solution in the glass vessel was initially one and a half inches above the tip of the experimental leaf. And as the leaf was growing nutrient solution was being added to keep the tip of the leaf one and a half inches below the solution level. And every third day fresh nutrient solution was added (to recoup the possible exhaustion). The plant being a completely sub-

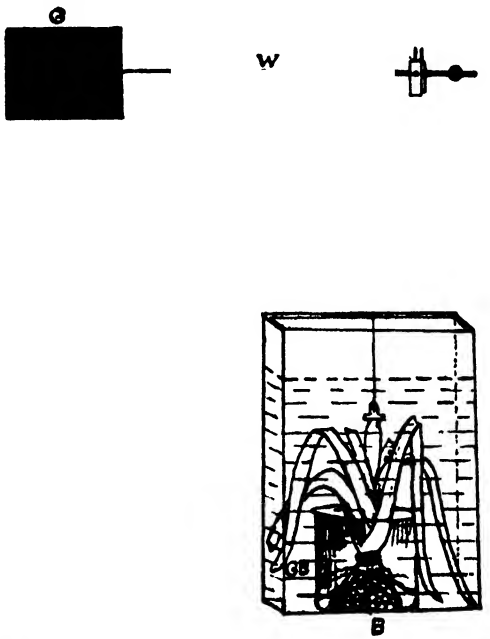


FIG. 1. Semi-diagrammatic representation of the complete arrangement for recording the longitudinal growth of a leaf of *Vallisneria spiralis*.
The stem of the specimen was fastened to a cylindrical glass flask B, loaded with lead shots. The specimen thus tied was placed in a glass beaker GB. The plant with the beaker was placed in a rectangular glass vessel R. The tip of the leaf to be experimented upon was clamped and was connected by glass links to the writing lever W which inscribes dots on the recording plate G. The level of water in the vessel R, was one and a half inches above the tip of the experimental leaf.
Successive dots are at intervals of 15 minutes.
Magnification employed was two and a half times.

merged water-plant, no additional oxygen was employed during the experiment. The temperature of the solution was found to vary through 4°C. The writing lever automatically inscribes dot on the recording plate G made to move to and fro by clockwork at an interval of 15 minutes. The point of the writer W, could be brought to the desired position within the plate by the adjustment of the fork on which the writer was pivoted. The plate was adjusted to move laterally through 6 inches in 24 hrs. The magnification employed in both these series of experiments was two and a half times only, since during the period of maximum growth a greater magnification would have carried the record outside the plate range.

The age of the particular specimen was calculated from the period which intervened between the date of appearance of the leaf on the axis and the date of experiment.

Simultaneous determinations of longitudinal growth of leaves of two specimens of the same age and length were made with two *Oscillating Recorders* having the same magnification and the same periods of dot intervals in the glass house in Knop's solution (*a*) with roots intact, and (*b*) fibrous roots cut off. Automatic diurnal records were taken.

Such determinations of longitudinal growth of two specimens (*a*) with roots intact, and (*b*) fibrous roots cut off were daily recorded from August 18 to August 30, 1937. Twelve records of each series (*a*) and (*b*) were taken noting the measurements of growth variations characteristic of different ages of the leaf.

The peculiarities observed in the variations of longitudinal growth of specimen (*a*) with roots, and (*b*) without roots will now be described in detail.

Experiment 1.

(*a*) *Longitudinal growth of a* 4 *days old specimen of a leaf of Vallisneria spiralis, with roots.*—In the record in Fig. 2(*a*) is shown the diurnal curve for 24 hours obtained on the 1st day from 6 P.M. of August 18 to 6 P.M. of August 19, 1937. It will be noted that the magnified growth during the 12 hours of night

p. 30, line 32, *insert* Plate 7 *after* Fig. 2(*a*)
p. 31, „ 15, „ Plate 7 „ Fig. 2(*b*)
p. 31, „ 28, „ Plate 7 „ Fig. 2(*b*)
p. 31, „ 31, „ Plate 7 „ Fig. 2(*a*)
p. 31, „ 32, „ Plate 7 „ Fig. 2(*b*)

p. 32, line 3, *insert* Plate 8 *after* Fig. 3(*a*)
p. 32, „ 6, „ Plate 8 „ Fig. 3(*b*)
p. 32, „ 19, „ Plate 8 „ Fig. 4(*a*)
p. 32, „ 23, „ Plate 8 „ Fig. 4(*b*)

from 6 P.M. to 6 A.M. is 15 mm. and that during the daytime from 6 A.M. to 6 P.M. is 22·5 mm. Thus the total magnified growth during 24 hours is 37·5 mm., the magnification being two and half times the total actual longitudinal growth in 24 hours is therefore $\frac{37{\cdot}5}{2{\cdot}5} = 15$ mm.

It will be seen that the first part of the curve, i.e. from 6 P.M. to 6 A.M. is relatively flat and the magnified growth being 15 mm. the actual growth is 6 mm. and the latter part of the curve from 6 A.M. to 6 P.M. the magnified growth being 22·5 mm. the actual growth is 9 mm. Thus the rate of growth during the 12 hours of the daytime is one and a half times greater than that during the 12 hours of night.

(*b*) *Longitudinal growth of a* 4 *days old specimen of a leaf of Vallisneria spiralis, without roots.*—The longitudinal growth of the specimen without roots given in Fig. 2(*b*), for 24 hours is obtained on the 1st day from 6 P.M. of August 18 to 6 P.M. of August 19, 1937. It will be seen from the curve that in the 1st part of it, i.e. from 6 P.M. to 6 A.M. during the 12 hours of night, the magnified record is 7·5 mm. the magnification here also being the same, namely two and a half times, the actual elongation in 12 hours is therefore $\frac{7{\cdot}5}{2{\cdot}5} = 3$ mm. The latter half of the curve is relatively very steep, the magnified record being 30 mm. the actual elongation is 12 mm. Thus the total actual elongation during 24 hours is 15 mm. Hence the average rate of growth during the daytime in this case is 4 times as great as that during the night.

Comparing the diurnal longitudinal records obtained in Fig. 2(*a*) and Fig. 2(*b*) it is found that the total increase of longitudinal growth in 24 hours in both the cases is the same, the amount being 15 mm. But the average rate of growth during the day in Fig. 2(*a*) is 1·5 times as great as that during the night, whereas in Fig. 2(*b*) the average rate of growth during the day is as great as 4 times that during the night.

Following the procedure adopted in Experiment 1(*a*) and Experiment 1(*b*) the observations were carried on from day to day and records taken till the leaf became fully mature on its attaining the age of 15 days.

EXPERIMENT 2.

(*a*) *Longitudinal growth of a* 5 *days old specimen of a leaf of Vallisneria spiralis, with roots.*—The next record of the series is reproduced in Fig. 3(*a*). The rate of growth during the night is 7 mm. and that during the day is 10 mm.

(*b*) *Longitudinal growth of a* 5 *days old specimen of a leaf of Vallisneria spiralis, without roots.*—The record is given in Fig. 3(*b*).

The actual rates of growth during the night and the day are 6·5 mm. and 12·5 mm. respectively.

EXPERIMENT 3.

(*a*) *Longitudinal growth of a* 6 *days old specimen of a leaf of Vallisneria spiralis, with roots.*—The record being similar is not reproduced here. Actual rates of growth during the night and that during the day are 8·5 mm. and 11 mm. respectively.

(*b*) *Longitudinal growth of a* 6 *days old specimen of a leaf of Vallisneria spiralis, without roots.*—The record being similar is not reproduced here. The growth elongation during the night is 9·5 mm. while that during the day is 14·5 mm.

EXPERIMENT 4.

(*a*) *Longitudinal growth of a* 7 *days old specimen of a leaf of Vallisneria spiralis, with roots.*—This record is reproduced in Fig. 4(*a*). The actual rates of growth during the night and the day are respectively 9·5 mm. and 12·5 mm.

(*b*) *Longitudinal growth of a* 7 *days old specimen of a leaf of Vallisneria spiralis, without roots.*—This record is given in Fig. 4(*b*). The respective rates of growth during the night and the day are 12·5 mm. and 17 mm.

EXPERIMENT 5.

(*a*) *Longitudinal growth of a* 8 *days old specimen of a leaf of Vallisneria spiralis, with roots.*—The record being of a similar nature to the previous one is not reproduced here. The rate of growth during the night was 10·5 mm. while that during the day was 14 mm.

(*b*) *Longitudinal growth of an* 8 *days old specimen of a leaf of Vallisneria spiralis, without roots.*—The record being similar to Fig. 4(*b*), Plate 8, is not given here. The rate of growth during the night and that during the day were 16 mm. and 19 mm. respectively.

EXPERIMENT 6.

(*a*) *Longitudinal growth of a* 9 *days old specimen of a leaf of Vallisneria spiralis, with roots.*—The record is given in Fig. 5(*a*), Plate 9. The respective rates of growth during the night and the day are 11·5 mm. and 14 mm. It will be seen from the curve that the growth activity begins to increase with the advancement of age but here when the leaf becomes 9 days old the activity is seen to be maximum, the total elongation during 24 hours being 25·5 mm. And the average rate during the day is 1·2 times as great as that during the night.

(*b*) *Longitudinal growth of a* 9 *days old specimen of a leaf of Vallisneria spiralis, without roots.*—The next record is reproduced in Fig. 5(*b*), Plate 9. The actual rates during the night and the day are 18 mm. and 21 mm. Here also as in the specimen with root the growth activity, as can be seen from the curve, is maximum, when the leaf became 9 days old ; the total elongation being 39 mm.

EXPERIMENT 7.

(*a*) *Longitudinal growth of a* 10 *days old specimen of a leaf of Vallisneria spiralis, with roots.*—The record being of a similar nature to that in Fig. 5(*a*), Plate 9, is not reproduced here. The growth elongation during the night was 10·5 mm. while that during the day was 12·5 mm.

(*b*) *Longitudinal growth of a* 10 *days old leaf of Vallisneria spiralis, without roots.*—The record being similar to Fig. 5(*b*), Plate 9, is not given in this case. The rate of growth during the night and the day was equal, being 15 mm.

EXPERIMENT 8.

(*a*) *Longitudinal growth of an* 11 *days old specimen of a leaf of Vallisneria spiralis, with roots.*—The record is represented

in Fig. 6(*a*), Plate 9. The actual rates of growth during the night and the day are 9 mm. and 10 mm. respectively.

(*b*) *Longitudinal growth of an* 11 *days old sepcimen of a leaf of Vallisneria spiralis, without roots.*—The record is reproduced in Fig. 6(*b*), Plate 9. The actual rates of growth during the night and that during the day are 10 mm. and 11·5 mm. respectively.

EXPERIMENT 9.

(*a*) *Longitudinal growth of a* 12 *days old specimen of a leaf of Vallisneria spiralis, with roots.*—Record being similar to Fig. 6(*a*), Plate 9, is not reproduced here. The respective elongations during the night and the day were 6 mm. and 8 mm.

(*b*) *Longitudinal growth of a* 12 *days old specimen of a leaf of Vallisneria spiralis, without roots.*—Record being similar to Fig. 6(*b*), Plate 9, is not given here. The rates of growth during the night and the day were 6·5 mm. and 8 mm. respectively.

EXPERIMENT 10.

(*a*) *Longitudinal growth of a* 13 *days old specimen of a leaf of Vallisneria spiralis, with roots.*—The record is reproduced in Fig. 7(*a*), Plate 10. The rates of growth during the night and that during the day are 3·5 mm. and 5·5 mm. respectively. Thus from the record in Fig. 7(*a*), Plate 10, it will be seen that the growth activity is further diminished.

(*b*) *Longitudinal growth of a* 13 *days old specimen of a leaf of Vallisneria spiralis, without roots.*—This record is given in Fig. 7(*b*), Plate 10. The respective rates of growth during the night and during the day are 3 mm. and 5·5 mm. Here also the growth activity has undergone a further diminution with the increasing age of the leaf.

EXPERIMENT 11.

(*a*) *Longitudinal growth of a* 14 *days old specimen of a leaf of Vallisneria spiralis, with roots.*—The record obtained was of a similar nature to Fig. 7(*a*), Plate 10, and is not reproduced here. The respective elongations during the night and the day were 2 mm. and 3 mm.

(*b*) *Longitudinal growth of a* 14 *days old specimen of a leaf of Vallisneria spiralis, without roots.*—The record taken being similar

to Fig. 7(*b*), Plate 10, is not reproduced. The relative rates of growth during the night and the day were 1·5 mm. and 3 mm. respectively.

EXPERIMENT 12.

(*a*) *Longitudinal growth of a* 15 *days old specimen of a leaf of Vallisneria spiralis, with roots.*—This, the last record of the series, is reproduced in Fig. 8(*a*), Plate 10. In the record in Fig. 8(*a*), Plate 10, the growth activity as will be seen has come almost to a stop. The elongation during 24 hours is only 1 mm. The rates of growth at night and at day are 0 mm. and 1 mm. respectively.

(*b*) *Longitudinal growth of a* 15 *days old specimen of a leaf of Vallisneria spiralis, without roots.*—This, the last record of rootless series, is represented in Fig. 8(*b*), Plate 10. From the curve it can be seen that the growth activity has practically ceased. The elongation during 24 hours is only 0·5 mm.

The results of the foregoing experiments on the variation in longitudinal growth of a leaf of *Vallisneria spiralis* (*a*) with roots are given in Tables I and II, and (*b*) without roots are given in Tables III and IV.

TABLE I.

Diurnal variation of longitudinal growth during the life cycle of the leaf of a specimen with roots of Vallisneria spiralis. (*Specimen* 1.)

Days of observation.	Longitudinal growth in mm. 6 P.M. to 6 A.M.	Longitudinal growth in mm. 6 A.M. to 6 P.M.	Total elongation in 24 hours in mm.	Average rate for 24 hours in μ per sec.
1	6·0	9·0	15·0	0·17
2	7·0	10·0	17·0	0·19
3	8·5	11·0	19·5	0·22
4	9·5	12·5	22·0	0·25
5	10·5	14·0	24·5	0·28
6	11·5	14·0	25·5	0·29
7	10·5	12·5	23·0	0·26
8	9·0	10·0	19·0	0·21
9	6·0	8·0	14·0	0·16
10	3·5	5·5	9·0	0·10
11	2·0	3·0	5·0	0·04
12	0·0	1·0	1·0	0·01
Mean	7·0	9·2	16·2	0·18

It will be seen from the above table that the longitudinal growth of leaf of a *specimen with roots* of *Vallisneria* 4 days old, on the first day of observation was 15 mm. On the next day growth elongation was 17 mm. After that when the leaf became 6 days old the elongation was 19·5. In this way the growth activity of the leaf began to increase gradually with the advancement of the age of the leaf, till on the 6th day of observation when the leaf became 9 days old the elongations became maximum being 25·5 mm. After this the elongation went on decreasing rapidly till on the 15th day of the age of the leaf when it practically came to a stop.

The following curve (Fig. 9) gives a graphical representation of the relation between the diurnal longitudinal growth and the age of the leaf of a specimen with roots.

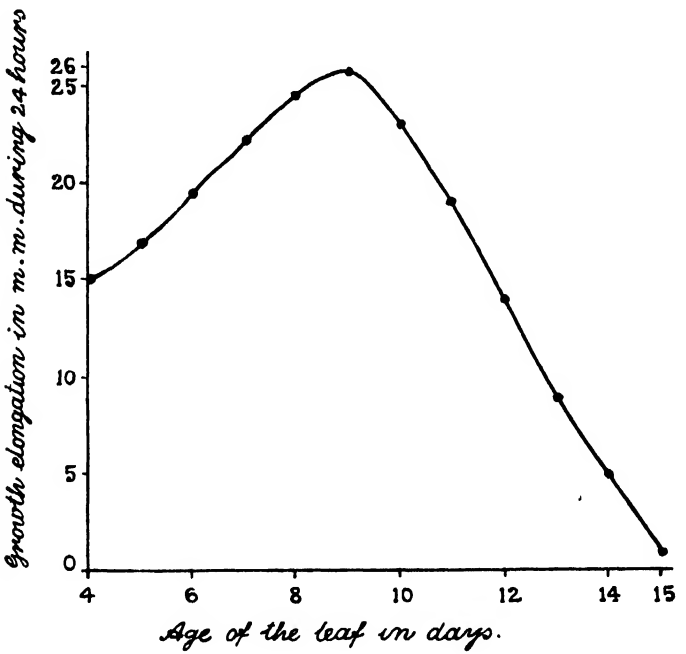


FIG. 9. Graphical representation of the relation between diurnal longitudinal growth with the age of the leaf, of a specimen with roots. The ordinate represents the diurnal longitudinal growth in mm., while the abscissa indicates the age of leaf in days.

It is to be seen in this connection that in the first few days the rate of growth went on increasing gradually with increasing age of the leaf. When the leaf became 9 days old, the rate of growth became maximum. Subsequently with increasing age of the leaf, the rate of growth began to decrease rapidly till on the 15th day when it practically came to a stop.

Confirmatory results obtained with a duplicate specimen of *Vallisneria spiralis, with roots*, is given in Table II.

TABLE II.

Diurnal variation of longitudinal growth during the life cycle of leaf of a specimen with roots of Vallisneria spiralis. (*Specimen* 2.)

Days of observation.	Longitudinal growth in mm. 6 P.M. to 6 A.M.	Longitudinal growth in mm. 6 A.M. to 6 P.M.	Total elongation in 24 hours. in mm.	Average rate for 24 hours in μ per sec.
1	4·0	6·0	10·0	0·11
2	5·0	7·0	12·0	0·13
3	6·5	8·5	15·0	0·17
4	7·5	9·5	17·0	0·19
5	9·0	11·0	20·0	0·23
6	10·0	12·0	22·0	0·25
7	8·0	10·0	18·0	0·20
8	6·5	7·5	14·0	0·16
9	5·0	6·5	11·5	0·13
10	3·0	4·5	7·5	0·08
11	2·0	2·5	4·5	0·05
12	0·0	0·5	0·5	0·005
Mean	5·5	7·3	12·8	0·14

A summary of results obtained in the previous experiments of the variation in longitudinal growth of the leaf of *Vallisneria spiralis, without roots*, is given in the following table:—

TABLE III.

Diurnal variation of longitudinal growth during the life cycle of a leaf of Vallisneria spiralis, *without roots.* (*Specimen* 3.)

Days of observation.	Longitudinal growth in mm. 6 P.M. to 6 A.M.	Longitudinal growth in mm. 6 A.M. to 6 P.M.	Total elongation in 24 hours in mm.	Average rate for 24 hours in μ per sec.
1	3·0	12·0	15·0	0·17
2	6·5	12·5	19·0	0·22
3	9·5	14·5	24·0	0·27
4	12·5	17·0	29·5	0·34
5	16·0	19·0	35·0	0·40
6	18·0	21·0	39·0	0·45
7	15·0	15·0	30·0	0·34
8	10·0	11·5	21·5	0·24
9	6·5	8·0	14·5	0·16
10	3·0	5·5	8·5	0·09
11	1·5	3·0	4·5	0·05
12	0·0	1·5	1·5	0·01
Mean	8·4	11·7	20·1	0·22

It will be seen from the above table that the longitudinal growth of leaf of a specimen of *Vallisneria, without roots*, 4 days old, on the first day of observation was 15 mm. On the next day when the leaf was 5 days old the elongation became 19 mm. In this way the elongation began to increase rapidly everyday and until the leaf was 9 days old when the growth elongation attained its maximum, becoming as great as 39 mm. Henceforth this activity began to diminish very rapidly and until the leaf became fully mature on its fifteenth day and there was almost complete cessation of growth.

The following curve (Fig. 10) gives a graphical representation of the relation between the diurnal longitudinal growth and the age of the leaf of a specimen of *Vallisneria spiralis, without roots*:—

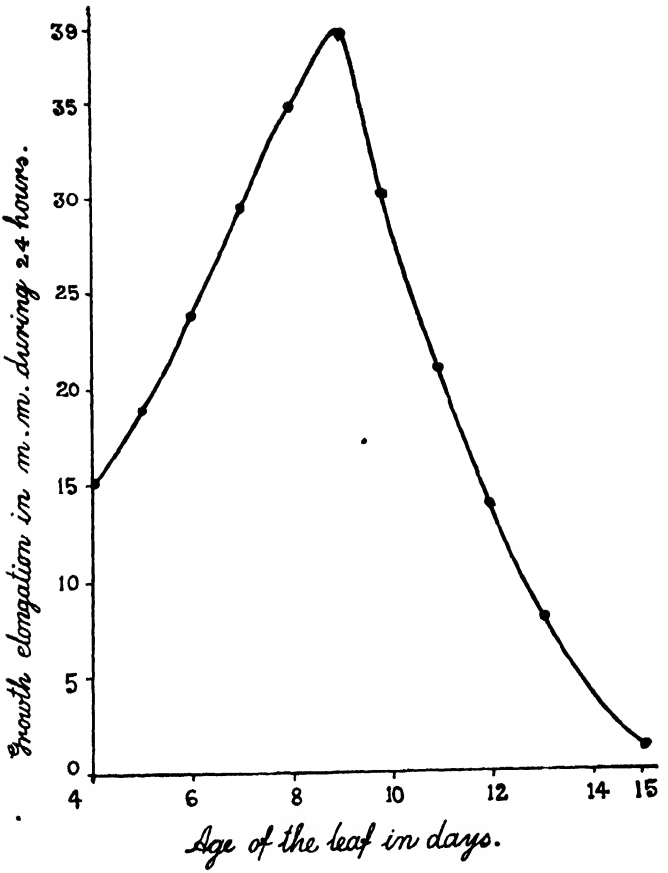


FIG. 10. Graphical representation of the relation between diurnal longitudinal growth with the age of the leaf of a specimen without roots. The ordinate represents the diurnal longitudinal growth in mm., while the abscissa indicates the age of leaf in days.

It is to be seen from the above curve that the rate of longitudinal growth, began to increase very rapidly for first five days. On attaining the age of 9 days the leaf showed its maximum rate of growth. Afterwards the rate of growth is found to decrease very rapidly with maturity of the specimen till on the 15th day of the age of the leaf the rate practically came to a stop.

Confirmatory results obtained with a duplicate specimen of *Vallisneria spiralis, without roots*, is given in Table IV.

TABLE IV.

Diurnal variation of longitudinal growth during the life cycle of a leaf of a specimen of Vallisneria spiralis, *without roots.* (*Specimen* 4.)

Days of observation.	Longitudinal growth in mm. 6 P.M. to 6 A.M.	Longitudinal growth in mm. 6 A.M. to 6 P.M.	Total elongation in 24 hours. in mm.	Average rate for 24 hours in μ per sec.
1	2·5	8·5	11·0	0·12
2	4·5	9·5	14·0	0·16
3	6·0	11·5	17·5	0·20
4	8·5	12·5	21·0	0·24
5	11·5	12·5	24·0	0·27
6	13·0	14·0	27·0	0·31
7	11·5	12·5	24·0	0·27
8	8·5	8·5	17·0	0·19
9	5·0	7·0	12·0	0·13
10	3·5	4·5	8·0	0·09
11	2·0	3·0	5·0	0·04
12	0·0	0·5	0·5	0·005
Mean	6·3	8·7	15·0	0·17

Discussion.

Dr. Jost in his lectures on Plant Physiology,[7] observed that 'numberless correlations make their appearance if members be *isolated*, if leaves, branches or roots of plants be separated off and prevented from rapid decay by appropriate artificial means. . . . . The root can give rise to shoots, the shoot to roots, the leaf to both shoot and root. The normal plant gives us no hint of this power, and yet that power must have been latent in it; the inter-relationships of the members only must have prevented the individual organs from exhibiting all the capacities which they possess'. Hill, Overholts and Poff in their book have remarked that[8] 'removal of the terminal parts of stems, for example, stimulates to growth lateral buds which normally would remain dormant . . . . . If the flower buds of tomato plants are pinched off as they appear, the plant is stimulated to renewed vegetative growth. On the other hand, if the fruits are allowed to develop, vegetative growth is

checked'. In our paper on 'Modification of vital activity after inflorescence in Mimosa'[9] it has been shown how the removal of flowers has stimulated the moto-excitability of the pulvinus of *Mimosa pudica*.

By adopting special methods and taking utmost care to keep the plant alive after removal of the roots it was found that the removal of roots stimulates the growth of the leaf.

Now from the data obtained from Table I, it will be seen that the longitudinal growth-variation of leaf specimens of *Vallisneria, with roots intact*, in culture solution is completed in the course of about a fortnight. The total amount of growth elongation during its life cycle is found to be 194·5 mm. The average rate of growth during 24 hours is 16·2 mm.

And from the data obtained from the Table III, it will be seen that the longitudinal growth-variation of leaf specimens of *Vallisneria, with fibrous roots cut off*, in culture solution is also completed in about the same period. The total amount of growth elongation during its entire life cycle is however found to be 242·0 mm. The average rate of growth in the course of 24 hours is thus 20·1 mm.

Thus it has been found that in the case of a specimen with roots cut off the total longitudinal growth is greater compared with that of a specimen with roots. The percentage increment of longitudinal growth in the case of *specimens without roots* during its entire life cycle is as great as 24 per cent.

Summary.

The present investigation was undertaken to ascertain the variation in longitudinal growth of leaves of *Vallisneria spiralis* in culture solution (*a*) with roots, and (*b*) without roots.

The specimen *with roots* showed progressive increase in longitudinal growth with the advancement of the age of the leaf, till on the 6th day of observation when the leaf was 9 days old; and the rate of growth attained the maximum activity. The rate of growth during the day was greater than that during the night. After that there was a gradual diminution of the rate of growth till on the 15th day of the age of the leaf the growth-elongation practically came to a stop. The longitudinal growth during

the day in the entire life cycle was greater than that during the night.

The specimen without roots exhibited rapid increase in longitudinal growth with the advancement of the age of the leaf till the age of the leaf was 9 days, when the rate of elongation attained its maximum. When the leaf was 10 days old the rate of growth during the night and the day was found to be equal. Then as the leaf became fully mature, the activity began to diminish till on the 15th day when the growth became practically arrested. The rate of growth during the day in its entire cycle was greater than that during the night excepting on the day when the leaf was 10 days old.

I take this opportunity of expressing my grateful thanks to the late Sir J. C. Bose and to Dr. D. M. Bose for the encouragement and helpful criticism which have been extended to me throughout this investigation.

References.

[1] Botany—J. Ben Hill, Lee O. Overholts and Henry W. Popp, p. 129, 1936.
[2] Water Plants—Arber Agnes, p. 204.
[3] Sauvageau, C. (1891).
[4] Pond, R. H. (1905).
[5] Brown, W. H. (1913).
[6] Das, S. C. and Palit, B. K.—Variation in longitudinal growth of a water plant during its life cycle. Trans. of the Bose Res. Inst., Vol. XI, pp. 9–27, 1935-36.
[7] Jost, Ludwig—Lectures on Plant Physiology, p. 252, 1907.
[8] Botany—J. Ben Hill, Lee O. Overholts and Henry W. Popp, p. 207, 1936.
[9] Das, S. C. and Palit, B. K.—Modification of Vital Activity after Inflorescence in *Mimosa pudica*. Trans. Bose Res. Inst., Vol. IX, pp. 8–34, 1933-34.

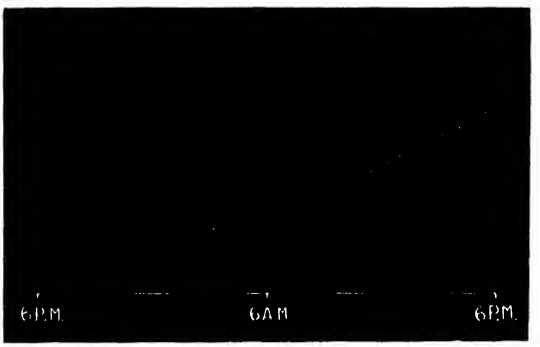


FIG. 2(*a*). Record of longitudinal growth of a specimen of a leaf of *Vallisneria spiralis* with roots when 4 days old.

The abscissa represents the hours of day and night, the ordinate representing growth elongation. The growth at any hour is found from the vertical distance of the curve from the point in the base representing the hour.

FIG. 2(*b*). Record of longitudinal growth of a specimen of a leaf of *Vallisneria spiralis* without roots when 4 days old.

The abscissa represents the hours of day and night, the ordinate representing growth elongation. The growth at any hour is found from the vertical distance of the curve from the point in the base representing the hour.

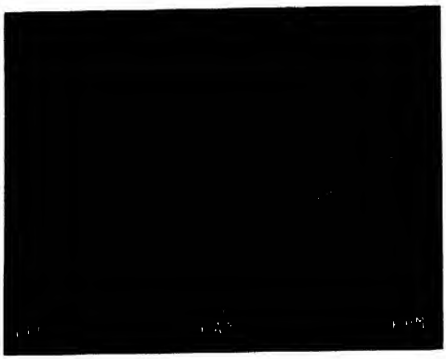


FIG. 3(*a*). Record of growth of a specimen of a leaf of *Vallisneria spiralis* with roots when 5 days old.

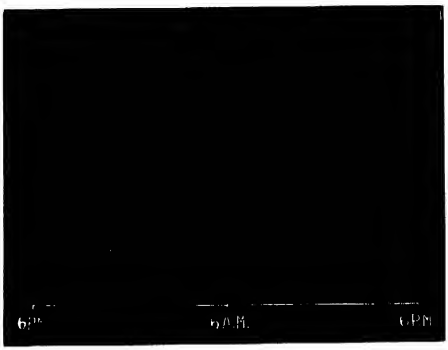


FIG. 4(*a*). Record of growth of a specimen of a leaf of *Vallisneria spiralis* with roots when 7 days old.

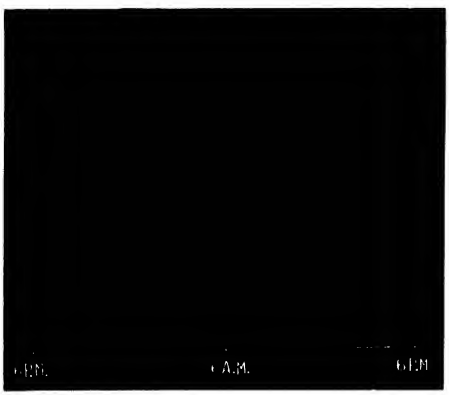


FIG. 3(*b*). Record of growth of a specimen of a leaf of *Vallisneria spiralis* without roots when 5 days old.

FIG. 4(*b*). Record of growth of a specimen of a leaf of *Vallisneria spiralis* without roots when 7 days old.

FIG. 5(*a*). Record of growth of a specimen of a leaf of *Vallisneria spiralis* with roots when 9 days old.

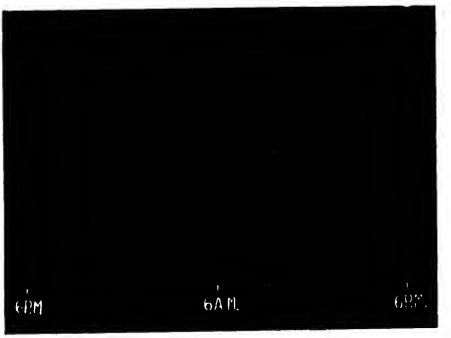


FIG. 6(*a*). Record of growth of a specimen of a leaf of *Vallisneria spiralis* with roots when 11 days old.

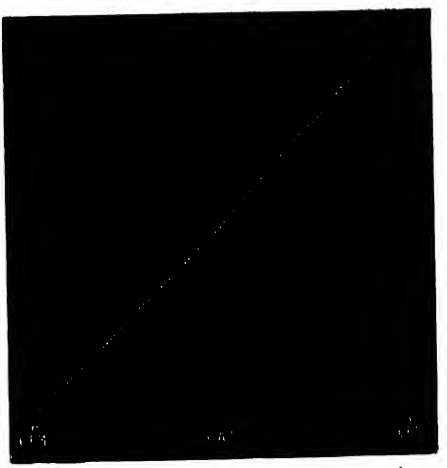


FIG. 5(*b*). Record of growth of specimen of a leaf of *Vallisneria spiralis* without roots when 9 days old.

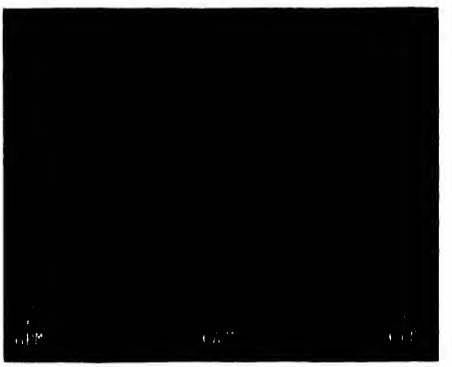


FIG. 6(*b*). Record of growth of a specimen of a leaf of *Valisneria spiralis* without roots when 11 days old.

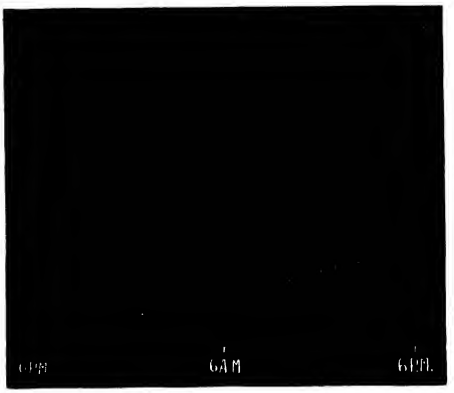


FIG. 7(*a*). Record of growth of a specimen of a leaf of *Vallisneria spiralis* with roots when 13 days old.

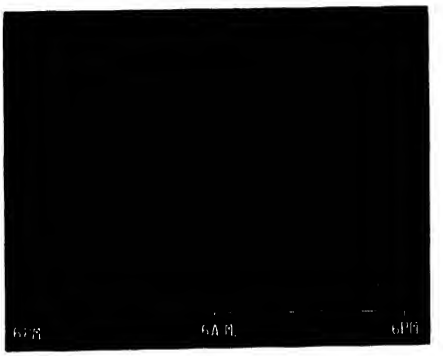


FIG. 8(*a*). Record of growth of a specimen of a leaf of *Vallisneria spiralis* with roots when 15 days old.

FIG. 7(*b*). Record of growth of a specimen of a leaf of *Vallisneria spiralis* without roots when 13 days old.

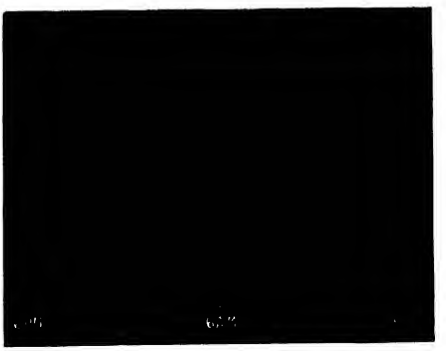


FIG. 8(*b*). Record of growth of a specimen of a leaf of *Vallisneria spiralis* without roots when 15 days old.

IV. NUTRITION EXPERIMENTS WITH THE INDIAN FOODSTUFF *PISUM SATIVUM* (II).

By N. C. NAG and A. K. PAIN.

(*Received 20th September, 1938.*)

In our paper on the above subject published in a previous issue of the Transactions [1], we gave details with definite results of observations till as late as March 1937. It was there noted that young albino rats, born of *Chhola* fed parents, when kept on *Pisum Sativum* (*Mutur*) diet from after the weaning period, failed in their power of procreation, indicating deficiency of Vitamin E. In contrast to the above observations, it was noted that *Chhola* fed (*Cicer arietinum, both Kabuli and Bengal*) albino rats from identical litters gave birth to young ones at regular intervals after attaining maturity.

Further experiments were continued during the latter part of 1937 and the earlier part of 1938. Examples of typical results obtained are detailed in this paper.

EXPERIMENTAL DETAILS AND RESULTS.

Male and female rats kept together on *Mutur* diet from after weaning period, and which had failed to give birth to young ones for eight months, were now given *Cicer arietinum* diet and observations continued. The results of this change in diet are detailed below:—

Cage No. 13.

Mutur fed rats for eight months were placed on *Chhola* diet on 1-4-37.	First litter of 11 was born on 26-7-37.

It will be observed that the power of procreation was effective after about three to four months on changed diet. Of

[1] Nag and Pain—Trans. Bose Res. Inst., Vol. XI (1935-36), pp. 91–97.

the litter of 11 young ones, seven are still alive on 4-12-37 and have attained full maturity.

Cage No. 14.

One female rat with one male rat fed on *Chhola* diet all along and giving birth to litters at regular intervals were placed on *Mutur* diet after the birth of their third litter.

The pair were placed on *Mutur* diet on 1-4-37.	Litter of 7 was born on 2-5-37. All the young ones died within half an hour of birth.

The birth of the litter just after about a month may be taken as the normal period of interval (normal period of gestation being 23 days or so). It is, however, important to note that none of the progeny survived. The effect of the *Mutur* diet was pronounced in this respect. There were no more litters born after this single one ; even this one litter may have been due to effectiveness of the previous *Chhola* diet still continuing.

Cage No. 15.

The experiment performed on rats in this cage was to investigate if a *Chhola* fed male rat could or could not effect impregnation of female rats fed on *Mutur*. The result is detailed below :—

One *Chhola* fed male rat was placed in the same cage with *Mutur* fed female rats on 1-4-37.	There were issues till the beginning of 1938.

It is to be noted that even if the male were normally fed, the deficiency in the female rats still prevailed.

A fresh series of observations was started with a view to examine the effect of mixed diet, and 50% *Chhola* mixed with 50% *Mutur* meal was first tried.

Cage No. 16.

Adult and fertile, now placed on 50% *Chhola* and 50% *Mutur* meal 19-11-37.

One male rat with —

(*a*) female ..	1st litter of 5 on 8-11-37.	2nd litter of 4 on 4-4-38.
(*b*) female ..	litter of 4 on 8-11-37.	No further.

It is to be remarked that none of the young ones survived.

Cage No. 17.

Experiments similar to the foregoing were started on the same date with another set of one male and two female rats, all adult and fertile before being placed on changed diet of 50% *Chhola* and 50% *Mutur*. Date of starting the observations—19-10-37.

One male with—

(*c*) female rat ..	litter of 7 on 11-11-37; 5 died within two weeks.	2 still alive on 3-12-37.
(*d*) female rat ..	This female did not give birth to young ones at all.	

The deficiency of the *Mutur* mixture was most pronounced in rat (*d*). The two young ones still alive on 3-12-37 (from the litter born to (*c*) on 11-11-37) weighed 15 grams each. These are rather underweights for the age when compared to young ones born of normally fed parents, actual weight in the latter case being 25 grams.

Cage No. 18.

Cage number 18 contained one male and three female rats all fertile adults, normally fed, now placed on *Mutur* diet on 19-10-37. One of the female rats died on 21-12-37. The two others did not give birth to young ones till January 1938.

SUMMARY.

That *Mutur* (*Pisum Sativum*) is deficient in Vitamin E is further confirmed. Even a mixed diet of 50% *Cicer arietinum* (*Chhola*) is not able to rectify the deficiency. However, a complete change of diet slowly restores the procreative power, and the progeny attain to maturity though they show underweight.

V. SOYA BEAN CULTIVATION AT FALTA.

By N. C. NAG, M.A., F.I.C.; H. N. BANERJEE, M.Sc.; and A. K. PAIN, M.Sc.

(*Received 1st August, 1938.*)

Soya Bean has been attracting a good deal of attention in India in recent years. It is claimed that the yield of the crop is considerable, so that when properly cultivated it should be a cheap foodstuff with high oil and protein contents, which are said to be easily assimilable. For a poor country the above considerations should be sufficient, unless of course there are other deterrent circumstances to be taken into account. It was with the intention of gaining first-hand knowledge on the above and other allied points that cultivation of Soya Bean was undertaken at the Falta Experimental Station, a rice producing centre in the 24-Parganas.

Our first supply of Soya Bean seeds was obtained in 1931 from Ottur, Muzafferpur. The Ottur farm belongs to Mr. S. K. Chakravarti and he was good enough to send us a small quantity of various seeds amongst which was Soya Bean, though these latter were rather small in size. Our preliminary experiments as regards raising of the plants from these seeds even so late as 1934, i.e. three years after, having proved successful, we began our systematic experiments with foreign seeds and seeds grown at Falta since 1935-36. The typical details are given below.

Suitable Season for Sowing.—It was found that plants could be grown at all seasons of the year. But plants grown at times other than between June to August were found to yield no crops or of generally very poor and neligible yield. If after planting there is excessive rain a high percentage (even so high as 50%) of the plants may get killed. But once the plants have risen to a height of two to three inches or so, they withstand heavy rain quite easily and thrive well.

Preparation of the plots for Soya Bean cultivation and Method of planting.—Two different procedures were adopted. One

was to grow the seedlings elsewhere and then transplant them at a suitable time. The well-ploughed field is arranged in rows of ridges at definite distances, the seedlings being planted also at definite noted distances. The intention of using this arrangement was to observe if there was any influence on the total yield of crop due to density of population (plant number per unit area).

The second method was to grow the plants on the ploughed field itself and then to arrange them on ridges so as to have as far as possible uniform distribution.

The only manure used was very light solutions of cowdung applied at the time of flowering, once a week for three to four weeks.

The actual results with dates for sowing and final harvesting, etc., are detailed below:—

Seeds sown during the earlier part of 1936 gave plants which fruited along with those sown during June.

Plot Number 1.—The actual size of the plot was $12\frac{1}{2}'$ by $27' = 337{\cdot}5$ sq. ft.

There were 15 ridges or rows, leaving 9 inches in between each row and some space along the two sidemost rows, thus covering the entire 12·5 feet.

In each row were planted at a distance of 9 inches in between Soya Bean seeds, there being thus altogether 33 seeds in each row, the total number being 500. There was practically cent per cent success in the germination and raising of the plants to maturity. The date of planting was 20th June, 1936. There was slight rain just a day after sowing and no heavy down pour for some time. The plants grew to a height of about 40 to 50 cm. by the early part of September, when flowers began to make their appearance. The seeds (pods) were ripe by the end of October and the plants were taken out of the field on the 1st of November, 1936. The actual weighing, after separation and collection of the seeds during the interval, was done on the 20th November, 1936.

Seeds from 492 plants were collected and weighed 3,490 gm. Yield per plant thus works out to be 7·09 gm. Weight of the

foreign seed sown per 1,000 was 69·46 gm., while the weight per 1,000 of the Falta product was 68·84 gm.

A comparison of the weight of the seeds sown and the amount obtained shows an yield of over 100, or more correctly 7·09/·069 = 102·0 times neglecting to take into account in this case the small percentage of ungerminated seeds.

Expressing the yield per acre, we get $\frac{14400 \times 3490 \times 3}{12{\cdot}5 \times 27}$ or say about 446,000 grams equivalent to 12 maunds per acre or 960 lbs. per acre.

It may be noted here that the density of the plants (or population) per acre in the present case was $\frac{500 \times 14400 \times 3}{12{\cdot}5 \times 27} =$ 64,000 Plants.

Plot Number 2.—The actual size of the plot was 40′ × 36′ = One-thirtieth of an acre.

The seeds were sown directly on the ploughed field on the 1st of July, 1937. After the seedlings had grown to a height of about 2 to 3 inches they were distributed so as to form 24 rows with 28 plants in each row. So that the total number of plants on the plot were 672.

As in the previous year, the flowering began in September and the final collection of the ripe seeds was finished by the end of November, 1937.

The total yield came to 8,926 grams. This works out to an average of 13·29 grams per plant. It may be recalled here that the average per plant in the previous year in Plot Number 1 was only 7·09 grams per plant.

A comparison of the weight of the seeds sown and weight of the total yield from those seeds shows that the apparent yield is better than that of last year, being 13·29/·069=193 times. The Falta grown seeds having an average weight of 0·0688 gm. per seed.

But expressing the yield per acre the result is only $\frac{8926 \times 14400 \times 3}{36 \times 40}$ = 267,780 grams per acre = 7·2 maunds nearly per acre, as compared with an yield of 12 maunds per acre in Plot Number 1.

This apparent discrepancy is explained when we take into account density of the plants per acre, which in the last case is only $672 \times 30 = 20,160$ per acre, while in Plot Number 1, it was no less than 64,000 or over three times the first.

Thus an increase of the space alloted to each plant by three times has nearly doubled the yield per plant.

Plot Number 3.—The actual size of the plot was $36' \times 8' = 288$ sq. ft. with only 144 plants evenly distributed. This was adjoining to Plot No. 2. The plants were raised exactly one month after those in No. 2, though the actual flowering and harvesting, etc. took place on the same date as in the case of No. 2.

The total yield for the Plot No. 3, was 1500 grams, or about 10·4 grams per plant. The somewhat late sowing was apparent also from the size of the plants, which were smaller but looked sturdier. The yield it will be noticed was about 25% less than that in Plot No. 2.

The yield per acre works out to be $1,500 \times 150 = 225,000$ grams or about 6·04 maunds per acre, even less than that in Plot No. 2. although the density of the plants was 21,600 plants per acre.

It may be remarked here that the effect of somewhat late planting was added to the defect in density, as compared with No. 1. Plot.

Summarising the results of the typical cases cited above it may be concluded that to get the best yield, the seeds should be sown between the middle of June and the middle of July or at the latest by the 1st week of August. This has been our experience in a number of other plot cultivations. Further to get the best yield per acre of land, it is better to sow the seeds at an average distance of 22 to 25 cms. apart (9 to 10 inches). More space gives better yield per plant, but reduces the population or density of the plants, and this defect may more than counter-balance the increased yield per plant.

Composition of the Soya Bean Seeds from Different Sources.

Along with the study as to the best results obtained regarding yield, the no less important question of the chemical compos-

tion of the locally grown seeds as compared with those of foreign source from which the Falta products were obtained was taken up. Recently we came to know of the Khadi Protistan Cultivation at Sodepur, about 15 miles to the north of Calcutta. We were shown over the farm by Messrs. Satis Ch. and Khitish Ch. Das Gupta. As regards cultivation and yield there was practically no difference from our own experience. Smaller sized seeds give better yield as regards total quantity per acre.

The following results are typical of several analyses of the different varieties :—

	Foreign seeds.	Falta grown Seeds (Mother seeds foreign).	Sodepur grown (Larger sized Mother seeds).
Weight per 1000 seeds in grams ..	69·46	68·4	73·24
Ash per cent ..	4·28%	5·56%	4·25%
Moisture (100°C.) ..	14·60%	13·65%	14·10%
Oil	12·45–13·00%	12·00–13·00%	13·03%
Protein	37·81%	37·76%	38·80%
Carbohydrates reducible to sugar ..	20·00%	20·00–21·00%	22·00%
	89·69%	90·97%	92·18%
Fibre, etc. by difference	10·31%	9·93%	7·82%

From an examination of the figures given above it would appear that the Sodepur product has certain apparent advantages. But it must also be remembered that these were grown from specially large sized seeds.

The percentage of ash in the seeds is appreciably greater in the Falta products. The presence of the mineral matter may not be a disadvantage. We give below the results of ash analysis.

Results of Analysis of Ash.

	Foreign.	Falta Grown.	Sodepur Grown.
SiO_2	0·70%	0·55%	0·50%
Fe_2O_3	1·70%	1·20%	1·30%
CaO	7·33%	11·36%	7·32%
MgO	9·16%	9·10%	9·49%
MnO	0·13%	0·23%	0·12%
K_2O, Na_2O (mostly K_2O)	46·00%	44·60%	48·10%
P_2O_5	33·32%	30·10%	30·30%
SO_3	1·75%	1·52%	2·01%
Cl	0·75%	1·25%	0·75%
Deduct for O ..	−0·16%	−0·25%	−0·16%
Total determined ..	99·68%	99·64%	99·73%

The main points that may be noted from the above results are that the Falta products give more ash and lime in the ash with somewhat lower percentage of alkali. However in this respect Sodepur products are on a par or even better than the foreign seeds we were dealing with. Quality of the products grown near Calcutta seems also to depend somewhat upon the locality where the Soya Bean has been cultivated. Nearness of Falta to the Ganges, the high chlorine content of the river water at certain seasons of the year makes its presence felt in the higher per cent of chlorine present in the ash. But speaking generally there is no deterioration in the quality of the beans when grown near Calcutta.

As regards the yield of oil, it was nearly the same in all the different samples. The oil in each case was of light yellow colour when freshly extracted. On exposure to light the colour gets slowly bleached. The oil which is of thin consistency to start with slowly gets viscid with exposure to air, and absorption of oxygen. The Iodine value of the different sample lay between 127 to 132. The saponification value lay between 187 to 188. The refractive index of the different samples was also the same and was 1·4715 at 31°C.

In estimating the quantity of carbohydrate convertible to reducing sugar, the method that gave uniformly consistent result was to treat the powdered Soya Bean with 10% HCl

in a stoppered pressure bottle dipped in a bath of boiling water for half an hour, and then to make the estimation in the usual manner after filtering and washing, etc.

Preliminary chemical colorimetric tests were not very definite as to the presence of Vitamins A, D, and C, beyond evidence of traces even after germination. As the physiological effects must be taken as the final criteria, albino rats were placed under observation with Soya Bean as food.

Physiological Observations.

If Soya Bean were to be given admixed with other foodstuff, the case would be complicated due to the influence of various other substances which might mask the effect of unmixed Soya Bean. We, therefore, decided to begin by giving unmixed Soya Bean and water to the subjects under examination as followed in our previous experiments[1]. Control experiments were also carried on side by side with *Cicer arietinum* diet, as the last had proved itself able to supply all the essentials for proper development and procreation of rats for the last eight years or so. In order to test the curative effect of Soya Bean diet, rats which were partially devitaminised by B.D.H. (vitamin free, fat free) preparation, were placed on Soya Bean diet and its effect compared with that produced by *Cicer arietinum*. Results of preliminary observation were published[2] by us some time back from which we quote the following:—

One litter of ten rats were divided into 5 cages A, B, C, D, and E, with two rats in each cage and fed as detailed below. Their weights on different dates were recorded.

Cage.	Dates and weights in grams.					
	30-3-36	5-4-36	12-4-36	19-4-36	24-4-36	3-5-36
(A) Chhola (common Cicer) Female ..	31	38·5	49	55·5	64	76·5
Male	31	41·5	55	70·5	84	96
(B) Soya Bean Female	35·5	40·5	50·5	53·5	51	died.
Male	38·5	41·5	57	68·5	80·5	83

From the above record it will be seen that one of the rats fed on Soya Bean died after about two months. This particular case was an isolated instance and might have been due to the female subject being peculiarly defective. However in our further experiments on the same line there was no other case like this. Young rats grow fairly well on Soya Bean diet and water only, at least for four months; when they give litters of young ones also, similar to that given by chhola fed rats. But the rate of growth or gain in weight is generally less than in the case of chhola fed rats, which is greatest in the case of rats fed with Kabuli Chhola, Soya Bean fed rats coming close after common chhola.

The rats in cages C, D, and E were first devitaminised by giving them B.D.H. deficient food; and then the effect of changed diet was observed.

Cage.	Dates and weights in grams. 30-3-36	5-4-36	12-4-36	19-4-36	24-4-36	3-5-36
		Deficient food.			Diet changed to C. chhola.	
C	34	35	36·5	36·5	62·5	82
	34	35·5	38·0	38·5	66·5	87
		Diet changed to Soya Bean.				
D	32	30	49	50	54·5	56
	33	34·5	45 *	51·5	54·5	60·5
E	33	died	Diet changed to Falta grown K. Chhola.			
	34		53	71	81·5	92

One of the rats died under deficient food before change of diet could be effected. However the records show that under Soya Bean diet there was appreciable response, though the gain in weight was appreciably less than under chhola diet.

Long continued observations for over two years have convinced us that rats reared on Soya Bean diet alone and water,

* Misprint in Science and Culture as 55 should be 45.

have shorter length of life. Most of the rats die within eight to nine months. Litters born to them are weaker and seldom survive more than four months and seldom grow to maturity. Young ones born of Soya Bean fed parents, however, when reared on chhola or mixed diet grow normally and can rear young ones in their turn.

SUMMARY.

Summarising the results of our observation and experiments we come to the following conclusions:—

The best time for sowing Soya Bean in the Falta locality is between the 15th of June and end of July. Using no other manure than cow-dung, the only manure that is easily available to an Indian cultivator without cost, he can get an yield of about 100 times the amount of seed sown, and with proper density of plants, about 4 maunds per Bengal Bigha or 12 maunds per acre. The produce should under the circumstances be much cheaper than the rate at which this is being sold at present. The yield varies with density of the plants, more sparsely distributed plants give even so much as $\frac{27}{0{\cdot}07} = 385$ times the amount of seed sown. But what is gained this way, is lost in there being lesser number of plants per acre of land. We obtained the best results when the space given was about 0·66 sq. ft. per plant.

As regards the quality of the products grown near about Calcutta, there was not much difference from those of foreign import. Physiologically too they gave similar results. The presence of high percentages of edible oil and protein combined with a low percentage of carbohydrate in Soya Bean, militates against its value as complete food. Rats reared on Soya Bean alone have shorter span of life. A mixed diet, however, avoids this risk and Soya Bean may serve as a cheap foodstuff when properly balanced with carbohydrate and also probably with other sources of vitamins.

We take this opportunity of gratefully remembering the inspiring words of late Sir J. C. Bose which led us to undertake

this work and compare the quality of Soya Bean with that of some of our Indian pulses, particularly *Cicer arietinum.*

REFERENCES.

[1] Trans. Bose. Inst. Vol. XI.

[2] Nag, Banerjee and Pain,—Science and Culture Vol. I, pp. 779-780, June 1936.

VI. CULTIVATION OF *PHASEOLUS CALCARATUS Roxb.* AND SOME FEEDING EXPERIMENTS.

By N. C. NAG and A. K. PAIN.

(*Received 10th August, 1938.*)

Along with our supply of Soya Bean Seeds from Mr. S. K. Chakravarti of Ottur, Mozafferpur, we had some seeds which looked like *Phaseolus radiatus* var. *Shona Moong*. The seeds were, however, much deeper in colour and bigger in size. The seeds, obtained so far back as April, 1932, showed life even after four years when 10 per cent of them germinated in May, 1936. One of the plants showed an unusual growth covering an area of at least 64 sq. feet. We pruned it down to cover no more than 9 sq. feet in the middle of August, and almost immediately within ten days after pruning there was copious flowering. Beans began to form and by the end of October the seeds were fast ripening. The actual collection of the seeds was completed on the 20th November, 1936. The collected seeds weighed no less than 131 grams. The average weight of a single seed being 0.039 grams, the yield was over 3,000 times. Calculated on acre basis, the yield might be taken as coming to about 90,000 grams or over two maunds per acre even supposing we had not pruned down the area covered. Over and above the actual amount of seeds collected quite an appreciable quantity had fallen down from the plant before the final collection could be effected. This promising yield encouraged us to continue the cultivation with seeds thus collected from a single plant. Next season in July, 1937 we planted an experimental plot of one: thirtieth of an acre with 15 grams of the collected seeds. The yield from this source was 7,000 grams. The plants, were identified by Dr. K. P. Biswas of Royal Botanical Gardens as *Phaseolus calcaratus Roxb.* 6.[1]

The plants of 1937 were not so spreading as that of the previous year. Indeed the whole area of one-thirtieth acre was not even fully covered. The yield per acre comes even then

to 210 kilograms or say roughly over 5 maunds. In this connection it may be worthwhile to note that while the *p*H value of the soil of 1936 was 7, that of 1937 was acid to begin with and near about 5, which slowly reached the neutral state after liming. The soil for 1937 was one dug out from an old tank bed which was re-excavated.

Pulses of the *Phaseolus* class form daily articles of food in almost every home in India. In Bengal and Northern India *Phaseolus* forms a subordinate crop and is often sown for the purpose of green manuring. *Phaseolus Mungo* is 'sown broad cast in April or May at the rate of 40 lbs. of seed per acre and the plants are hoed into the land after 6 to 8 weeks'[2]. The presence of cyanogenetic glucosides have been mentioned by several authors and although caution has been advised no case of actual poisoning has been as yet attributed to these pulses.[3]

Sir George Watt[2] mentions that *Phaseolus radiatus* sown at the rate of 2½ to 3 seers per acre yielding in November 5½ to 7 maunds of seeds and 30 maunds of fodder. This works out to, at best, 100 times the seeds sown. Again *Phaseolus aconitifolius*, bir-mung, matti kalai, is mentioned as being reaped in November-December yielding 120 lbs. per acre for 1½ lbs. of seed sown—and this is regarded as a fair yield.

Analysing these figures with those obtained by us from our *P. calcaratus* cultivation at the Falta Farm of the Bose Research Institute there seems ample room for a fair trial being given to *P. calcaratus* cultivation, should the feeding experiments not definitely pronounce against this pulse. *Phaseolus radiatus* var. *Shona Moong* was shown to be approaching *Cicer arietinum* in its nutritional value. And *P. calcaratus* in appearance resembles *P. radiatus* and the yield having been found to be so promising it was decided to examine the seeds obtained from our 1936 and 1937 cultivation. The present paper thus deals with some preliminary observations on the physical, chemical and some physiological properties of *P. calcaratus*.

Results of Physical and Chemical Examination.

Phaseolus calcaratus seeds are darker in colour than the *Shona moong* seeds and in size also much bigger, approaching

in the latter respect those of *P. aconitifolius* var. Bengali Black Mashkalai.

	Weight per 1,000 seeds.	Specific gravity.
P. radiatus ..	20·35 grams average.	1·32 average.
P. calcaratus ..	35 „ „	1·35 „
P. aconitifolius ..	44·53 „ „	1·38 „

Picked seeds of *P. calcaratus* weigh more than the average ones and may go to so high a value as that of the Black Mashkalai. Indeed an average of 39 grams for 1,000 seeds is not uncommon.

Chemical Composition of P. calcaratus.

Moisture	8·47%
Ash content	4·16%
Oil by Ether Extraction	1·80% or less.
Carbohydrate reducible to sugar by HCl treatment	53·21%
Protein (N. determination)	20·65%
Fibre	9·13%
Total determined	97·42%
Pigments and other soluble matter ..	2·58% by diff.

Mineral constituents in the Ash.

Insoluble in HCl	1·19%
Fe_2O_3	1·19%
CaO	15·37%
MgO	3·58%
P_2O_5	15·55%
Alkali (Mostly K_2O)	47·28%
SO_3	5·32%
Cl	0·93%
MnO	0·08%
Total determined	90·49%

The undetermined portion consists mostly of CO_2.

It may be remarked here that in composition *P. calcaratus* is like the other pulses and shows a high percentage of alkali, and P_2O_5 in the mineral portion.

The total quantity of *P. calcaratus* seeds collected at the Falta Farm was only 10,500 grams (7,000 grams from 1/30th acre and another 3,000 to 3,500 grams collected from plants which grew from seeds which had fallen off the 1936 single plant). It was, however, decided to start some physiological experiments by feeding albino rats following the procedure generally adopted by us and described in more detail in our other published paper. We give here only the results obtained within as short a space as possible.

Physiological Observations.

The first observation was started with a pair of albino rats born on the 25th of December, 1937. The parents of this pair were normally fed with *Cicer arietinum*. It so happened that both the young ones were females. They were put on whole meal *P. calcaratus* diet on the 29th of January, 1938. Their weights were 36 and 39 grams being quite normal for the age of about 5 weeks. The results of regular weighings taken at intervals are recorded in the following table:—

The weights are given in grams under different dates.

Dates	29/1/38	4/2/38	10/2/38	28/2/38	14/3/38	22/3/38	29/3/38
Rat 1	36	42	50·5	77	86	91	93
Rat 2	39	43	48	76·5	84	90	92

Weighings were taken on other and more frequent dates, but they showed no particular variation in the regular increment of the body weight of the rats; they are not given here to avoid taking too much space without in any way bringing out any new feature.

It will be seen that the two rats had reached the maturity stage as evidenced from the weights, 90 to 95 grams being the normal for females, as also by their age. They were now placed

in a cage with a male rat and all of them placed on whole meal *P. calcaratus* diet. The increase in weight continued and attained to 110 and 108 grams on the 20th of April, 1938. In fact, the females were carrying young ones.

Rat 1 gave birth to a litter of 4 on the 27th of April, 1938, that is just about a month after the female was placed with the male. The usual gestation period is 23 days or so.

Rat 2 gave birth to a litter also of four on the 28th of April, 1938, that is just a day after the first.

The normal time taken by rats to give their first litter is generally between 110 to 120 days. In the case of the *calcaratus* fed rats also the first litters appeared in about 4 months time.

Fresh Set for Observation.

In the above cases the two females were placed with a male which had been fed on *Cicer arietinum* diet until the time when this was placed with the *calcaratus* fed females. In the fresh set of experiments, one pair of rats, one male and one female, born on the 16th of February, 1938 were placed after their weaning period on the 19th of March, 1938 in Cage No. 30 on whole meal *calcaratus* diet. Weights of the rats on different dates are recorded below:—

Cage No. 30.

Dates	..	19/3/38	29/3/38	9/4/38	19/4/38	7/5/23
Male	..	28	33	53	53	69
Female	..	27·5	35·5	45·5	61	87

The first litter was born on the 4th of June, 1938. The litter however, failed to survive beyond a few days. It will be noticed that the male of the pair was rather weak and unusually under-weight.

Devitaminisation Experiments.

The next set of three female rats from the same litter as those in Cage No. 30 were placed in Cage No. 40 on fat free

vitamin free diet, until deficiency diseases appeared. They were then placed on *calcaratus* diet to see if this diet made any improvement in their condition. The records are detailed below:—

Cage No. 40—B.D.H. *Deficient Food.*

Dates ..	19/3/38	26/3/38	29/3/38	
(*a*) Female Rat	29·5	28	28·5	Eyes affected.
(*b*) „ „	30·5	30	30	„ „
(*c*) „ „	28·5	29·5	29	„ „

The eyes were affected in all the three rats and they were losing weight: sure signs of danger which might soon take them beyond the stage of curing. Indeed the Rat (*a*) died before reaching the age of maturity even though the diet was changed to whole meal *calcaratus* on the 29th of March 1938, almost immediately after devitaminisation was evident. The result of diet change is recorded below:—

Cage No. 40—Calcaratus diet after B.D.H.

Dates ..	29/3/38	2/4/38	9/4/38	19/4/38	7/5/38
Rat (*b*) with affected eyes ..	30	Eyes cured. 38	45.5	65	85
Rat (*c*) with affected eyes ..	29	35	48 Eyes curing.	68 Cured.	86

The above details bring out the fact that *calcaratus* diet not only cured the devitaminised rats but also that the rats by their weights showed that they had attained maturity, by the 7th of May, 1938.

These rats were then placed on the 10th of May, 1938 with the male rat from Cage No. 30, the one reared on *calcaratus* from after weaning period.

The first litter to Rat (*b*) was born on the 1st of June, 1938 but all the young ones failed to survive the weaning period.

To Rat (*c*) there was no progeny till the middle of June, 1938, when this paper is being drawn up.

P. calcaratus *and* P. radiatus *compared.*

Side by side with the experiments carried on with *calcaratus* feeding experiments with *radiatus* var. *Shona Moong* were being repeated as the latter had already been found to have very high food value. The following records are interesting:—

Dates	30/3/38	19/4/38	7/5/38
Rat A male *P. radiatus* fed	25	47	79
Rat B female *P. radiatus* fed	27	45	67

	19/3/38			
Rat 1 male *Calcaratus* fed	28	32	53	69
Rat 2 female *Calcaratus* fed ..	27·5	34·5	61	87

In the above case the feeding was commenced on the 19th of March, 1938 in the case of *calcaratus* and on the 30th of March, 1938, that is 11 days later, in the case of *Shona Moong* and naturally enough the older subjects may be expected to show greater gain in weight. However, the comparative figures are interesting and justifies further observation both as regards cultivation possibilities and food value, of *Phaseolus calcaratus.*

Summary.

Phaseolus calcaratus Roxb. has been cultivated starting with a single seed. The yield per acre as also the number of times calculated on quantity of seed sown basis are both promising.

The physiological experiments though as yet not sufficiently numerous to lead to any definite conclusions, the chemical

composition and the preliminary experiments showing curative effects in disease brought on by deficiency diet justify further work.

REFERENCES.

[1] Roxburgh.—Flora Indica, Ed. 1874, p. 555.

[2] Sir George Watt.—The Commercial Products of India, Ed. 1908, pp. 225, 880.

[3] Sir George Watt.—Quotations from—

Dunstan and Henry—Proc. Royal Soc., 1903, 72, 281 and Agri. Ledg., 1902, 2; Jour. Board Agri., 1908, XIV, pp. 722–731.

Church—Food Grains of India, 1886, p. 155.

Leather—Agri. Jour. India, 1906, I, pt. III. p. 224. Cyanogenesis in Plants.

Additional.—Transactions of the Bose Research Institute.

Nag and Banerjee.—'On oil Soluble Vitamins in *Cicer arietinum*.' Vol. IX, pp. 89–118.

VII. EFFECT OF INDIVIDUAL AND MIXED MANURES ON RICE CULTIVATION.

By ANIL K. PAIN, M.Sc.

(*Received 15th July, 1938.*)

Falta, in the 24 Paraganas, is an important centre for rice cultivation. Generally the rice lands get submerged during the rains and take on silt and the cultivators do not use any extraneous manures. During the dry season, the Ganges water enters through natural canals and the lands and particularly the canal sides get deposits of salt, which, however get washed off by the sweet water with the advent of rains.

The present experiments were carried out at the Experimental Research Station of the Bose Institute at Falta. The land itself has been made somewhat higher in level than the canal water level. However there was arrangement for proper supply of water by means of pumps.

The seeds.—The seeds selected for these experiments were those locally known as *Baktulsi* of finer variety. The seeds were sown on the 8th of July, 1937 on a small unmanured piece of land which was kept watered for facilitating good germination. It took just three days for germination which was very successful. Five seers of seeds were actually sown. When the seedlings were 27 days old they were transplanted to prepared plots. The quantity actually required for our special experiments was indeed only about one-tenth of the whole quantity that was grown. The surplus amount was taken away by the local cultivators, still leaving a certain amount for a side cultivation.

Preparation of the plots.—An unmanured highland was selected and was divided into six plots, each measuring one hundred and twentieth (1/120th) part of an acre. These plots were well ploughed and cleared. The ploughing was done thrice over at an interval of about a fortnight before the transplantation of the seedlings. The actual dates of ploughing were 30th June, 16th July, and 4th August, 1937.

Manuring.—Of the aforesaid six plots five were manured and the *sixth* was kept unmanured as *control.* The actual manuring was carried out in the following manner. Two of the Plots A and B, were given complete manure containing Nitrogen, Potash, and Phosphorus. Three of the other Plots, C, D, and E were treated individually and separately with the three different manures. The plots were numbered and treated in the following manner :—

Plots	A, B	Complete manure—4 lbs. Calcium Phosphate, 3 lbs. Ammon. Sulphate, and 2 lbs. Potash Chloride.
Plot	C . .	Nitrogen—3 lbs. Ammon. Sulphate.
Plot	D . .	Phosphorus—4 lbs. Calcium Phosphate.
Plot	E . .	Potash—2 lbs. Potash Chloride.
Plot	F . .	Control—No manure.

Phosphate and Potassium Chloride in Plots A, B, D, and E were given on the 15th of July, 1937, Ammonium Sulphate to Plots A, B, and C was given on the 4th of August, 1937.

Transplantation.—The seedlings were transplanted to the respective plots on the 4th of August, 1937 when they were 27 days old and with an average height of 27 inches. In each of the plots *single seedlings* were planted separately. The plots measured 12′ by 30′ each. There were 16 rows of 30 plants in each row. Each separate plot was protected by high ridges all around to prevent washings entering from other plots. The plots were watered whenever they showed tendency to dry.

Control Soil analysis.—An analysis of the soil from the selected land was carried out before the experiments were started. Samples of the soil from different parts of the land were taken and mixed so as to obtain a fair sample. The mineral constituents were determined. The results are given in Table I.

TABLE I.

Analytical Results of Air Dry Soil.

Loss on ignition	8·97%	
SiO_2 and insolubles	74·18%	(Concentrated HCl).
Al_2O_3	7·03%	
Fe_2O_3	4·90%	(Iron total as ferric).
CaO	1·33%	
MgO	0·10%	
K_2O	0·70%	
P_2O_5	0·49%	
TOTAL	97·70%	
Undetermined	2·30%	

Total Nitrogen content in the air dry soil was determined by the standard Kjaedahl Method and found to be 0·098%.

Analysis of the seedlings.—On the day the seedlings were transplanted, 500 carefully washed seedlings were taken and dried to constant weight in a steam oven. A portion of the dry material was carefully burnt to ash at comparatively low temperature. The mineral constituents of the ash were determined and the results are expressed per 100 grams of dry matter.

Weight of dry matter per 100 seedlings	11·2 grams.
Average Nitrogen content	1·15%
Ash	18·36%

TABLE II.

Mineral Constituents in Ash (Seedling) from 100 grams of dry matter.

SiO_2	12·41
Fe_2O_3	0·11
P_2O_5	0·50
CaO	0·32
MgO	0·18
K_2O	4·21

It will be observed that 12·41/18·36 or about two-thirds of the ash consists of Silica, and more than one-fifth consists of Potassium Oxide, while the quantity of P_2O_5 is also an important constituent.

Examination of plants at different periods of growth.—In order to find out the variation in composition of the plants at different periods of their ages, samples were taken out and examined, the summary of the results obtained are given below. The actual sampling was done by taking 16 plants at random one from each row and from different parts of the plots. The first sample was collected 30 days after the seedlings were transplanted. During collection of samples the number of tillers in each plant was also noted. The average number of tillers and the average height of the plants were also taken. From the data thus obtained a comparative idea of growth of the plants can be formed.

Plots A and B were two complete manure plots. The results here given were obtained as average from the two samples.

TABLE III.

Sample 1, Collected on 4-9-37.

Plot No.	Treatment.	* Average number of tillers per plant.	Average height of plant in inches.	Dry matter per 100 plants.
A, B	Complete manure	19·4	42	156 grams.
C	Nitrogen	23	48	259
D	Phosphorus	10·0	36	87
E	Potash	11·9	36	89
F	No manure	11·6	36	98

From the above figures it may be noted that the growth of the plants is greater in the Nitrogen treated plot, viz. Plot C.

* Details regarding total counts are given in Table III(*a*).

Next comes the complete manure Plots A and B. The plants in the three other plots D, E, and F showed very nearly the same figures.

Tiller Counting.

Sample 1, Collected on 4-9-37.

TABLE III(*a*).

Row No.	Complete Manure Plot A.		Complete Manure Plot B.		Nitrogen Plot C.		Phosphate Plot D.		Potash Plot E.		Control Plot F.	
	Plant No.	No. of Tillers.	Plant No.	No. of Tillers.	Plant No.	No. of Tillers.	Plant No.	No. of Tillers.	Plant No.	No. of Tillers.	Plant No.	No. of Tillers.
1	5	19	5	24	18	3	4	16	13	28	8	8
2	9	4	17	20	16	25	7	11	9	14	12	11
3	8	22	28	20	1	21	1	6	5	12	19	12
4	19	24	15	21	5	25	24	9	20	12	3	9
5	25	17	9	17	8	40	10	14	18	12	7	14
6	16	19	3	24	24	20	22	12	4	10	5	15
7	4	16	16	17	18	18	14	12	6	9	9	13
8	28	25	2	25	28	24	12	15	22	13	11	8
9	3	16	18	15	6	28	4	9	21	15	22	13
10	10	12	24	13	8	24	25	12	25	12	28	8
11	8	23	28	33	26	24	13	11	11	15	22	11
12	24	35	17	17	7	19	23	9	9	8	11	12
13	16	14	1	13	4	19	4	10	12	12	5	12
14	1	23	3	24	18	26	19	11	16	15	16	14
15	7	23	5	21	17	18	28	10	1	11	23	13
16	23	25	18	20	13	18	20	11	21	7	19	13

The chemical examination of the plants from the different plots was also carried out, and the results are given in Table IV.

TABLE IV.

Mineral constituents per 100 grams dry matters. (Sample 1.)

	Average of A and B	C	D	E	F
Ash ..	26·58	20·66	25·80	20·05	17·66
SiO_2 ..	20·66	15·14	19·97	14·21	12·89
Fe_2O_3 ..	0·38	0·23	0·19	0·15	0·11
P_2O_5 ..	0·45	0·45	0·78	0·46	0·40
CaO ..	0·29	0·35	0·28	0·30	0·26
MgO ..	0·24	0·17	0·20	0·19	0·18
K_2O ..	3·31	3·44	3·37	4·21	3·22

It will be seen that in the case of seedlings the quantity of SiO_2 in the ash was about two-thirds of the total (see above in Table IV). After 30 days growth the quantity of ash per 100 grams of dry matter has increased in quantity as also the proportion of SiO_2 in the ash. While in the seedlings to start with the proportion was 2 is to 3 in the present case after 30 days the proportion is more like 3 is to 4. Another point which may be noted is that the quantity of P_2O_5 is greatest in the Phosphate treated plants, similarly in the case of Potash treated plants K_2O is appreciably greater.

Flowering of the plants.—Flowering started after two and a half months from the date of transplantation of the seedlings. The actual dates are given below :—

TABLE V.

Dates of Flowering.

A	15-10-37
B	17-10-37
C	17-10-37
D	19-10-37
E	18-10-37
F	19-10-37

Sample 2 was collected on the 5th of November, 1937. The crop had just begun to mature. The entire specimens were examined as before and the results are given in Table VI.

TABLE VI.*

Sample 2, Collected on 5-11-37.

Plot No.	Average height of plant in inches.	Average number of tillers per plant.	Dry matter in grams per 16 plants.	Weight of rice in gram per 16 plants.
A and B	72	14·3	857·6	391
C ..	72	12·0	673	308
D ..	63	8·2	476·3	211
E ..	66	11·2	636	383
F ..	63	9·2	497	271

An examination of Table III will show that the dry matter was the greatest in amount in plants of Plot C which was the Nitrogen treated plot. In Table VI, however, the greatest amount of dry matter is found in plants reared with complete manure, i.e. in plants of Plots A and B. Further the plants from other plots have also shown improvement in this respect, comparatively speaking. It may further be remarked that plants grown on potash Plot E, have shown distinct improvement in dry matter weight.

* On comparing the number of tillers given in Table III, Sample 1, and those given in Table VI, Sample 2, a diminution in the number of tillers per plant is noticeable. This is due to all the tillers not surviving till the last.

Tiller Counting.

Sample 2, Collected on 5-11-37.

TABLE VI(*a*).

	Complete Manure Plot A.		Complete manure Plot B.		Nitrogen Plot C.		Phosphate Plot D.		Potash Plot E.		Control Plot F.	
Row No.	Plant No.	No. of Tillers.	Plant No.	No. of Tillers.	Plant No.	No. of Tillers.	Plant No.	No. of Tillers.	Plant No.	No. of Tillers.	Plant No.	No. of Tillers.
1	9	12	19	16	6	7	17	7	17	16	12	8
2	16	15	11	15	17	8	6	6	4	9	17	13
3	14	15	24	9	24	10	14	10	12	12	12	9
4	20	30	7	12	19	16	12	8	18	10	..	10
5	6	15	14	11	15	11	26	11	9	9	4	8
6	3	14	16	21	2	13	20	8	27	18	27	9
7	5	21	22	13	6	19	6	9	21	11	25	8
8	27	14	3	16	14	8	22	8	17	11	18	8
9	9	16	12	10	10	12	12	12	14	13	4	10
10	20	8	14	13	17	14	10	6	18	9	19	7
11	19	6	10	15	4	11	17	9	10	8	4	7
12	7	12	3	14	26	25	24	9	8	10	14	10
13	14	21	24	12	17	9	18	9	24	11	2	9
14	24	18	11	14	16	19	12	11	21	10	6	13
15	21	13	20	10	9	10	2	9	9	13	10	11
16	19	7	19	18	5	15	24	6	5	10	16	18

The final harvesting was done on the 28th of November, 1937, three and a half months after transplantation of the seedlings. Sixteen plants from each plot were taken and the paddy separated. The weight of paddy from each batch of sixteen plants

was noted and the total yield was hence calculated and noted as so many maunds (maund=about 80 lbs.) per acre. Table VIII is a statement of the yield in the different plots.

TABLE VII.

Mineral constituents of the ash of Sample 2 plants (parts per 100 grams of dry matter).

	A and B	C	D	E	F
Ash ..	17·21	16·42	15·96	15·77	15·81
SiO_2 ..	13·70	14·09	12·42	12·07	12·45
Fe_2O_3 ..	0·24	0·12	0·18	0·18	0·17
P_2O_5 ..	0·40	0·38	0·60	0·37	0·38
CaO ..	0·23	0·20	0·23	0·25	0·23
MgO ..	0·18	0·14	0·18	0·24	0·17
K_2O ..	1·47	0·89	0·40	2·14	1·42

An examination and comparison with former results show that the ash content and hence the mineral matter contents have appreciably decreased with age of the plants. It would seem that with maturity of the plants the mineral constituents are transported elsewhere.

TABLE VIII.

Yield of Paddy and of Straw.

Plot No.	Weight in grams of Paddy of 16 plants.	Straw from 16 plants.	Calculated Paddy in mds. per acre.
A and B ..	516	1,550	48
C ..	408	1,184	37·8
D ..	274	1,024	25·4
E ..	490	1,009	45·4
F ..	368	779	34·1

Table VIII brings out the fact that the yield of *paddy as also of straw is greatest* in the case of complete manure Plots

A and B, and is about one and one-half times greater than that in the control (unmanured) Plot F. The Potassium Chloride treated plot gave an yield of paddy very nearly equalling the complete manured plots, but was poorer in straw. In the Ammonium Sulphate treated Plot C the growth of the plants was very marked during the earlier stage of growth but the rate of growth slowed down towards the later and nearing maturity. The yield of paddy was slightly better than that in the control, while the straw yield was appreciably greater. The Phosphate treated plot showed an actual decrease in paddy yield when compared with the control, though the straw yield was somewhat better.

It may be worth while mentioning that the local cultivators who use no manure and depend upon the yearly silt deposit get an yield in best years of 30 to 35 maunds of paddy per acre. Our own yield using complete manure or even only Potassium Chloride is about one and one-third or one and one-fourth time better.

SUMMARY.

Rice plants have been grown in experimental plots each measuring 30 feet by 12 feet, i.e. 1/120th of an acre under unmanured control and manured conditions. The manures used were Ammonium Sulphate, Calcium Phosphate, and Potassium Chloride, singly and as mixture. Towards the earlier stages response was found to be greatest as regards dry matter formation in Ammonium Sulphate treated plots. But in the final matured stage the yield of paddy and dry straw is best in complete manured plots. Next best response was found in Potassium Chloride manured plots. A transportation of the mineral matter was noticed in the mature stage from the plant portion to elsewhere. Compared with the yield obtained by the local cultivators our yield was much superior except in the case of mere Phosphate treatment.

I take this opportunity of expressing my grateful thanks to the late Sir J. C. Bose for his constant encouragement during the progress of the investigation and to Prof. N. C. Nag for encouragement and helpful criticism all through.

VIII. ON THE CONSTITUTION OF CLERODIN, THE ACTIVE BITTER PRINCIPLE OF CLERODENDRON INFORTUNATUM. PART II.

By H. N. BANERJEE.

(*Received 1st September, 1938.*)

In a previous paper [1] a partial formula for Clerodin, the active bitter principle of *Clerodendron Infortunatum*, was suggested on the basis of some derivatives then prepared. The molecule of Clerodin was found to contain a pair of double linkages, a free OH group and an acetyl grouping. Further confirmatory evidence regarding the above observations were afforded by the following experiments.

Catalytic hydrogenation of Clerodin in a mixture of cyclohexane and acetic acid by means of Platinum catalyst (Adams and Shriner's) showed that after 18 hours' contact with shaking, one molecule of hydrogen was absorbed by one molecule of Clerodin.

Quantitative estimation of active hydrogen by the method of Tschugaeff and Zerevitinoff shows the presence of one single active hydrogen atom in the Clerodin molecule. By the action of freshly prepared Grignard Reagent on Clerodin in dry iso-amyl ether only one molecular proportion of methane was evolved and collected. It is evident that the acylable OH group contains this hydrogen atom.

Although Clerodin could not be reduced by means of aluminium amalgam in absolute ether, it could be easily reduced by means of zinc dust and acetic acid. The di-hydro Clerodin thus obtained was found to be identical with the product obtained by catalytic hydrogenation of Clerodin.

A large number of experiments have been performed with the object of degrading Clerodin to simpler products in order to determine the nature of the carbon skeleton present in the Clerodin molecule.

By the action of 10% sulphuric acid the acetyl group of the Clerodin molecule is lost and a product identical with the di-hydroxy compound obtained from Clerodin by hydrolysis with alcoholic KOH was obtained. It is a di-hydroxy compound and does not give any colour reaction with ferric chloride or Millions Reagent.

By the action of cold concentrated HCl the acetyl group is lost along with the loss of a molecule of water. Simultaneously an atom of chlorine enters the molecule. The halogen derivative ($C_{11}H_{15}OCl$) thus formed did not seem to contain any free OH group because it could not be acylated or benzoylated.

By fusion with KOH under milder condition an amorphous product, insoluble in all the ordinary organic solvents, was obtained from Clerodin along with some acetic acid. Preliminary analysis showed that the amorphous powder was not pure. By subjecting this to more drastic fusion an acid (M. Wt. 208) was obtained. No other definite product could be isolated.

Distillation with soda-lime under carefully controlled conditions yielded a yellowish green viscous product from which no crystallisable solid matter could be obtained.

When subjected to dry distillation at a high temperature Clerodin undergoes decomposition into a deep brown viscous mass which solidified on long standing. At 250°/20 mm. a few drops of a greenish yellow mobile liquid with a distinct acid reaction distilled over and was found to contain amongst other products traces of acetic acid.

On pyrogenic reduction with zinc dust in a current of H_2, Clerodin yielded a large volume of gaseous products and a yellowish green mobile liquid of high boiling point which could be purified by repeated distillation under high vacuum and finally through the picrate. Analysis showed this to be a hydrocarbon. It is unsaturated, absorbs bromine and reacts with fuming nitric acid with explosive violence, forming a nitro-derivative. Unfortunately the yield of this hydrocarbon was extremely small and it was not possible to investigate it further.

Along with the above another highly fluorescent thick oil of still higher boiling point was obtained in very small quantity. This product could not be made to yield a picrate and did not give constant analytical data after repeated attempts at purification.

Attempts were made to detect whether Clerodin molecule contains any benzene or fused benzene nuclei by the method of Ruzicka[2]; although H_2S was evolved in profuse quantity no trace of any pure aromatic hydrocarbon could be isolated from the reaction products. Dehydrogenation with sulphur was therefore repeated under milder conditions in toluene solution by the application of the method of Morton and Horvitz[3]. Under the condition of the experiment there was no reaction whatever between sulphur and Clerodin, as Clerodin could be recovered unchanged at the end of the experiment.

As dehydrogenation with sulphur under varying conditions did not prove to be fruitful another attempt at dehydrogenation of Clerodin was made with selenium. After boiling with selenium for 18 hours at about 200°C and subjecting the mixture to vacuum distillation a greenish yellow mobile liquid was obtained. This was found to be identical with the liquid hydrocarbon obtained from Clerodin by pyrogenic reduction with zinc dust in a current of hydrogen.

When Clerodin was oxidised with potassium permanganate in neutral solution an aromatic monobasic acid ($C_{13}H_{16}O_4$) crystallising in plates M.P. 265°C d. was obtained from the reaction product.

On treatment with nitric acid Clerodin gives carbon-dioxide, oxalic acid and small amount of a yellow micro-crystalline product, which is a nitro-derivative. (M. P. = 206°C.)

Oxidised with Kiliani chromic acid solution Clerodin is partly oxidised into an acid identical with the acid $C_{13}H_{16}O_4$, obtained by oxidation with potassium permanganate.

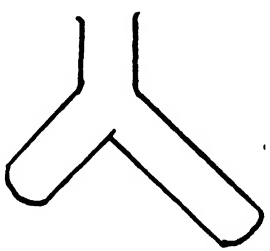
In the previous paper, the possibility of Clerodin as an anthelmintic drug was indicated by its toxicity to earth-worms. This property has been further corroborated by experiment with human thread-worms obtained from the stool of a young child. *Oxyuris vermicularis* put into a 1% colloidal suspension

of Clerodin in a solution of bile salts made slightly alkaline ($p_H = 7{\cdot}8$) with sodium carbonate were killed in five minutes, whereas the controls kept in alkaline solution of bile salts alone could be kept alive for a much longer period.

Some experiments were also performed in order to see how much of the Clerodin fed to animals were absorbed by their system. Experiment with rats and rabbits showed that the ingested Clerodin was eliminated through the stool. No trace of Clerodin could be detected in the urine (clarified by lead-acetate and de-leaded by sodium sulphate), so that there was no indication of absorption of the drug within the system. By means of a suitable device the urine and stool of the experimental animals were made to accumulate separately into two different chambers. These were removed every 12 hours and examined. In these experiments the colour reaction, which developed with acetic acid and ammonia as mentioned in the previous paper, was utilised for the detection of Clerodin in the stool and urine.

Experimental.

Di-hydro Clerodin:—0·02 g. of Clerodin was subjected to catalytic hydrogenation according to the absolute method of Smith [4]. in an apparatus prepared according to Smith's description with slight modification in the reaction bulb and in the compensation bulb. These bulbs consisted of a two-limbed vessel exactly similar to the reaction vessel used in the Zerevitinoff determination as represented in the diagram.



The Clerodin was put into the shorter limb and the solvent and catalyst in the longer one. In this arrangement during preliminary shaking for saturation of solvent with hydrogen

the solvent cannot splash into the substance. But during the actual hydrogenation of Clerodin, the contents of the two limbs could be very easily mixed together by simply tilting the apparatus. By this modification the danger, associated with 'vigorous shaking' for breaking the sample tube containing the substance to bring it in contact with the catalysing mixture, was avoided.

In the present case the solvent used for hydrogenation was a mixture of cyclohexane containing 10% recrystallised acetic acid. The mixture was previously saturated with H_2 at 3 to 4 atmospheres in presence of platinum catalyst. 20 c.c. of such solvent and 0·2 g. Pt. catalyst was used for the present hydrogenation. After 18 hours' contact with shaking the volume of hydrogen absorbed corresponded to 1·1 molecule of H_2 for one molecule of Clerodin.

The reaction mixture was filtered from the Pt., washed free of acetic acid and the light brown solution treated with charcoal and filtered. The dried cyclohexane solution was evaporated when crystalline plates free from bitterness were obtained. These melt at 115°C (with decomp.) with preliminary shrinking at 80°C.

Determination of active hydrogen in Clerodin:—This determination was carried out according to the method of Tschugaeff and Zerevitinoff[5]. 0·1 g. dry Clerodin was dissolved in 10 c.c. of well-dried amyl ether and allowed to react with an excess of freshly prepared Grignard Reagent. There was immediate evolution of methane which was collected in a Lunge Nitrometer by washing through brine. The volume of methane collected reduced to N.T.P. came to 10·3 c.c. Theoretically 0·1 g. Clerodin would liberate 10·09 c.c. of methane, calculated on the basis of one active hydrogen atom per molecule of Clerodin.

Reduction of Clerodin by zinc dust and acetic acid:—Zinc dust in small quantities was added to boiling solution of Clerodin (1 g.) in 25 c.c. glacial acetic acid, and the mixture heated for three hours. After filtering from unchanged zinc dust, the filtrate was evaporated to dryness under reduced pressure from the water-bath. A slightly brown gelatinous mass was obtained

which gradually solidified. It was dissolved in chloroform, washed free from adhering acetic acid, dried and evaporated to dryness. Crystalline plates were obtained from dioxane solution which were tasteless. It did not absorb bromine in chloroform showing that the double bond had been saturated. It melts at 114-115°C with decomposition.

Found C = 69·4%; H = 9·01%.

$C_{13}H_{20}O_3$ requires C = 69·64%; H = 8·93%.

A mixture of this substance with the product obtained by catalytic hydrogenation of Clerodin melted with decomposition at 115°C with preliminary shrinking at about 80°C, indicating their identity.

Action of dilute sulphuric acid:—Clerodin 1·2 g. and 10% H_2SO_4 (100 c.c.) were heated on a water-bath in a conical flask under reflux for two hours. Most of the crystals went into solution, and a drop of colourless oil was found floating above the clear colourless solution. On gradual cooling the floating oil solidified and was separated from the reaction product. It is bitter, and is found, after purification and recrystallisation from alcohol, to be unchanged Clerodin. The turbid solution deposited on cooling a tasteless brown amorphous product. The solution was cooled in ice and filtered. The clear filtrate did not show any rotation and did not reduce Fehling's solution after addition of excess of alkali. Tested by the Shaffer and Hartman micro method [6] no trace of reducing sugar was found.

The tasteless brown precipitate (0·2 g.) was crystallised from a large volume of absolute alcohol with charcoal and was obtained in plates M.P. 250°C. It contains free hydroxyl group and was identified as the di-hydroxy compound obtained from Clerodin by hydrolysis with alcoholic KOH.

Action of cold concentrated hydrochloric acid:—Finely powdered Clerodin crystals (2 g.) were added to 10 c.c. of well-cooled hydrochloric acid (sp. gr. 1·19). The crystals dissolved immediately to an intense pink coloured solution. After a short time precipitate began to appear. At the end of two hours the reaction product was poured into an excess of water, thoroughly stirred and filtered. The well-washed pink precipitate

was dried in the steam oven and kept in a vacuum desiccator over KOH for a month. The substance (0·6 g.) is soluble in glacial acetic acid but insoluble in all other ordinary organic solvents. It does not contain any free hydroxyl group as it did not react with phenyl-isocyanate and could not be acetylated or benzoylated. Boiling alcoholic KOH had no action on the substance. From glacial acetic acid it was obtained as a fine pink microcrystalline powder which does not melt below 360°C. Analysis shows that the molecule contains an atom of chlorine in its composition, and that it is produced from Clerodin by the loss of the acetyl group and also the elimination of a molecule of water in which process the free OH group present in Clerodin takes part.

The substance fuses above 360°C, takes fire and burns away with a sooty flame, without leaving any residue.

Found C = 66·6; H = 7·65; Cl = 17·62%.

$C_{11}H_{15}OCl$ requires C = 66·49%; H = 7·55%; Cl = 17·88%.

Fusion with potassium hydroxide:—Clerodin (5 g.) was gradually added to KOH (60 g.) and water (6 c.c.) at 120°C. Reaction took place with much frothing and the reaction mass was maintained at 200°C for half an hour when a granular precipitate separated out of the molten reaction product. The cooled melt was dissolved in a large volume of water when a light brown insoluble granular product (2 g.) settled down. This was filtered, washed with water and treated in glacial acetic acid. It was found to be insoluble in boiling glacial acetic acid and also in all ordinary organic solvents, as well as in molten camphor. It does not melt up to 360°C. Heated on the platinum foil it froths up, then chars and burns away without leaving any residue. Shaken in a solution of diazomethane in ether for several hours the substance remained unchanged. As its identification presented great difficulty, it was subjected to a further more drastic fusion with KOH.

1·2 g. of the powder was therefore intimately mixed up with 30 g. of powdered KOH and the mixture fused at 250–300°C for half an hour. A characteristic peculiar smell was emitted

and an insoluble red scum floated above the molten mass. On cooling, the scum solidified before the KOH did, and was mechanically separated out from the KOH melt. It dissolved easily in cold water into a deep brown solution from which HCl precipitated a brown sticky mass. This was dissolved in ether, washed, dried and the ether evaporated. A lac-like solid cake (0·53 g.) was obtained, highly soluble in cold acetone and alcohol but insoluble in water. After several purifications from boiling alcohol (charcoal) the substance was obtained as a deep brown vitreous brittle solid strongly adhering to the walls of the glass vessel. It was soluble in organic solvent into a deep brown solution. After neutralisation and formation of silver salt the molecular weight of the acid (taking it to be dibasic) was found to be 208. (M.P. 90-91°C.)

The aqueous alkaline filtrate, obtained from the first KOH fusion, after acidification and extraction with ether gave a small quantity of acetic acid only. Nothing could be obtained from the second KOH melt.

Soda-lime distillation of Clerodin:—10 g. of Clerodin crystals were intimately mixed with powdered soda-lime in large excess and the mixture heated in a dry pyrex tube over a soda-nitrate and potash-nitrate bath in a current of N_2 to drive off the evolved moisture. At about 250°C water began to collect in the receiver kept immersed in ice. After 10 to 15 minutes the receiver was changed and the temperature of the bath raised to 350°C and maintained there for three hours. Dense brown combustible fumes began to escape and a few drops of a viscous yellowish green oil collected in the receiver. The fumes dissolved in benzene into a yellow solution with a distinct green fluorescence. On raising the bath temperature no further distillate collected. On cooling, the oil gradually thickens and turns deep brown in contact with air. It has a peculiar characteristic odour somewhat resembling burnt rosin. It is highly soluble in ether, benzene and chloroform but sparingly in alcohol. With concentrated H_2SO_4 it gives a blood red colouration.

At 250°C/20 mm. the viscous brown product distilled over. But it did not solidify after several months and analysis proved that it was not a homogeneous product. Purification through

the picrate proved ineffectual as it could not be made to yield a picrate from ether, alcohol, or acetone solutions.

Action of heat on Clerodin:—Clerodin (0·5 g.) was heated in a pyrex test tube at 170 to 175°C in an oil-bath. The substance melted. A delivery tube was attached to the mouth of the test tube, the free end of which was dipped in baryta water. The baryta did not turn milky until at 250°C. Although the substance turned deep brown, no sublimate or distillate could be collected at higher temperature.

Distillation under reduced pressure:—5 g. of Clerodin were distilled at 20 mm. in a small distilling flask, the receiver being kept immersed in a freezing mixture. At about 145°C/20 mm. the crystals melted and frothed up but at 250°C/20 mm. a few drops of a greenish yellow mobile liquid with a distinctly acid reaction collected in the receiver. The distillate which was of a disagreeable odour dissolved in chloroform and decolourised Bromine in chloroform. The resinous reaction product, left in the distillation flask, solidified on cooling into a deep brown vitreous solid with a bitter taste. It dissolved in alcohol and chloroform into a deep brown solution. No crystalline solid could be isolated from this.

Pyrogenic reduction with zinc dust:—An intimate mixture of 5 g. Clerodin and an excess of zinc dust was heated in an elongated pyrex bulb, arranged according to the method of Jacobs and Craig [7], over a KNO_3 and $NaNO_3$ bath in a current of pure dry hydrogen gas. At about 250°C the reaction began and a deep green mobile liquid, along with much water condensed in the receiver, kept immersed in a freezing mixture. The receiver was then changed and the temperature raised to 360°C, when a viscous brown oil distilled over. The temperature was raised a few degrees higher and the reaction was complete. Large volume of gaseous products were evolved throughout the course of the reaction. These did not condense in the freezing mixture. On cooling, the reaction product was broken, thoroughly powdered, and extracted in ether. A small quantity of the high-boiling viscous product was thereby obtained.

The brown viscous mass:—This product (0·3 g.) did not solidify on keeping in a vacuum desiccator for several weeks.

It has a strong smoky odour, dissolves easily in cold ether and acetone but sparingly in alcohol. The solutions are yellow with a deep blue fluorescence. With concentrated H_2SO_4 a blood red colouration is produced and it reacts with fuming HNO_3 with explosive violence. The substance could not be obtained sufficiently pure to yield any constant analytical data. It burns away with much sooty flame.

The greenish mobile liquid:—This product (0·4 c.c.) turns deep orange on standing in contact with air, reduces Bromine in chloroform very vigorously. The brominated solution rapidly turns deep blue on standing.

The picrate prepared from alcoholic solution is obtained in plates M.P. 131°C from absolute alcohol. This was decomposed by Na_2CO_3 to regenerate the liquid hydrocarbon.

Found C = 90·01%; 89·98%. H = 9·9%; 9·99%.

With fuming HNO_3 reaction took place in the cold with explosive violence. The nitrate crystallised from alcohol in needles melts with incipient decomposition at 88°C; N = 8·21%.

The quantity of the liquid was exhausted so that further characterisation was not possible.

Dehydrogenation of Clerodin:—

(*a*) The dehydrogenation of Clerodin was first carried out by Vesterberg's method[8]. By heating Clerodin (5 g.) with excess of sulphur at 200°C to 250°C there was copious evolution of H_2S and perceptible odour of mercaptan. The reaction product was distilled at 200°C/20 mm. and a deep brown viscous distillate was obtained. This did not solidify even after one month in a vacuum desiccator over strong H_2SO_4. It could not be made to yield a picrate. With cold concentrated HNO_3 a nitrate was formed which could be reduced with tin and hydrochloric acid and the product diazotised and coupled with alkaline B-naphthol to yield a beautiful scarlet dye. No further characterisation was possible as the yield was very small.

Dehydrogenation with sulphur was therefore carried out under milder conditions in order to get, if possible, a better yield of the final product. For this purpose 0·37 g. of Clerodin was boiled with its equal weight of flowers of sulphur in a small

flask in 20 c.c. toluene as solvent under reflux. The free end of the reflux condenser was attached to a bent glass tube which was passed into a wash bottle containing aqueous alcoholic lead acetate. The mixture was boiled over an oil-bath continuously for 115 hours. The lead acetate solution remained untarnished. Apparently no reaction had taken place. On cooling, monoclinic prisms of sulphur separated out of the reaction mixture. From the solution 0·17 g. of unchanged Clerodin was recovered. By evaporation of the solution under reduced pressure a tarry paste was obtained which was similar in nature to the pasty product obtained from Clerodin when it is heated alone for 4 hours at 125°C, or when its toluene solution is boiled alone for 100 hours.

(*b*) Dehydrogenation with selenium:—An intimate mixture of Clerodin (5 g.) and selenium (12 g.) was heated in a 200 c.c. long-necked pyrex flask fitted with a very long reflux condenser, over a paraffin-bath.

At about 210°C vigorous reaction began with evolution of noxious fumes and deposition of red flowers of selenium in the cooler parts of the tube. After the vigorous reaction had subsided, the molten reaction product was kept briskly boiling at 170°C for six hours. At this point a fresh quantity of 2 g. of selenium was added and the mass boiled at 170°C for 12 hours more. On cooling the reaction product was powdered and extracted with ether. Ether left a thick orange gum which was subjected to vacuum distillation at 170 to 200°C/20 mm. A deep green mobile liquid (0·2 c.c.) was collected which solidified at 10 to 11°C. This product, on further purification by redistillation under high vacuum at 150°C was found to be identical with the liquid hydrocarbon obtained by pyrogenic reduction of Clerodin by zinc dust in presence of H_2, both giving identical nitrate and picrate.

The thick orange pasty residue left in the distilling flask, solidified into a vitreous brittle solid with a strong odour of hydrogen selenide. No crystalline solid matter could be obtained from this.

Action of Oxidising Agents on Clerodin :—

(*a*) Neutral potassium permanganate :—Clerodin (5 g.) was dissolved in acetone (50 c.c.) and powdered $KMnO_4$ (4 g.) added with stirring and cooling. The product was then diluted with water (50 c.c.) and a rapid stream of sulphur dioxide passed through the solution. The clear colourless solution thus obtained, was then thoroughly extracted with ether. The ether was washed with water, dehydrated over anhydrous Na_2SO_4 and evaporated to dryness when a transparent gelatinous mass was obtained. This solidified into a glassy solid on standing for one month in a vacuum desiccator over H_2SO_4. It was crystallised from glacial acetic acid in hard plates M.P. 265°C (decomp.). It is insoluble in water, but soluble in alcohol. Heated on a platinum foil it froths up and then burns away with a sooty flame. Molecular weight of the substance obtained by the silver salt and lead salt methods, taking it to be monobasic, was found to be 230·4 and 237 respectively. The acid reacts with ethereal solution of diazomethane, but no solid substance could be obtained from the reaction product.

Found C = 65·92%; H = 6·89%.

$C_{13}H_{16}O_4$ requires C = 66·101%; H = 6·77%.

Molecular weight—236.

(*b*) Dilute nitric acid:—2·0 g. of the substance and 5 c.c. of nitric acid (d. 1·36) were taken in a boiling tube provided with a delivery tube that dipped in clear baryta water. The nitric acid solution was heated in a water-bath at 60°C. The baryta turned milky and gradually barium carbonate began to settle down. But after a short time nitrous fumes began to evolve. After four hours the reaction product was further heated for one hour on the boiling water-bath and the intense orange-coloured solution poured into an excess of water. Copious yellow precipitates were obtained. These were filtered, thoroughly washed with water and finally crystallised from acetone (charcoal) in yellow microcrystalline powder (0·31 g.). It is highly soluble in dilute alkalies and could be precipitated unchanged by re-acidification. It melts at 206°C with decomposition.

Found C = 61·22%; H = 7·14%; N = 7·23%.

The mother liquor left after separation of the nitro-compound was concentrated under reduced pressure. A deep orange solution was obtained.

This was extracted with ether. The dehydrated ethereal solution after evaporation left an orange viscous mass which on standing partially solidified into a beautiful crystalline mass. This was pressed against porous plate and the solid crystals were obtained almost pure in slender colourless needles. After recrystallisation, these were found to reduce acid $KMnO_4$ vigorously. The crystals were identified as oxalic acid via the calcium salt and also by determination of mixed melting point with a pure sample of oxalic acid. The orange gummy mass separated from the oxalic acid did not solidify after standing several months in a vacuum desiccator over H_2SO_4. It is highly soluble in water. Further characterisation was not possible as the quantity recovered was very small.

(*c*) Kiliani-chromic acid solution :—A solution of Clerodin (1·4 g.) in acetic acid (10 c.c.) was treated with 5 c.c. of kiliani-chromic acid solution. The solution was vigorously shaken for 5 minutes and the mixture diluted with water and left overnight. The green solution was then thoroughly extracted with chloroform. The well-washed chloroform solution was then shaken up with dilute Na_2CO_3 solution, then washed free from the carbonate, dehydrated and then evaporated to dryness.

From the Na_2CO_3 solution by adding an excess of cold dilute HCl an acid was precipitated (0·1 g.). It was insoluble in water but soluble in alcohol from which it crystallised in plates M.P. 265°C (with decomp.). It was found to be identical with the acid obtained from Clerodin by $KMnO_4$ oxidation. A mixture of the two did not show any lowering of the melting point.

The chloroform solution left a bitter gelatinous mass (0·7 g.) which gradually solidified. This after purification and crystallisation from 60% alcohol was found to be still bitter. After several crystallisations from alcohol (charcoal) the substance melts with decomposition at 117°C to 120°C. The

bitterness was persistent and the substance was found to be lævorotatory:

$$[\alpha]_D = -21.$$

Analysis showed that it was not a pure homogeneous product. It could not be made to react with ethereal diazomethane solution. It gave blood red colouration with concentrated H_2SO_4 and was highly contaminated with unchanged Clerodin.

Further work is progressing.

CONCLUSIONS.

The nature of the hydrocarbon obtained by zinc dust distillation of Clerodin and the behaviour of the nitro-compound obtained from the dehydrogenated Clerodin prove conclusively that the nuclear hydrocarbon of the Clerodin molecule has a cyclic structure and that the unsaturation is associated with the ring.

It is hoped that further experiments with large amounts of Clerodin will throw definite light on the structure of the Clerodin molecule, which has already been proved to possess the following structural formula:

$$C_{11}H_{14}\begin{cases} O.CO.CH_3 \\ OH \end{cases}$$

My thanks are due to Late Sir J. C. Bose and to Prof. N. C. Nag for the interest they have shown during the course of this investigation.

REFERENCES.

1 Banerjee, H. N.—Trans. Bose. Res. Inst., 11, 71, 1936.
2 Ruzicka.—Helv. Chim. Acta., 4, 505, 1921.
3 Morton and Horvitz.—J. Amer. Chem. Soc., 57, 1860, 1935.
4 Smith, J. H. C.—J. Biol. Chem., 96, 35, 1932.
5 Tschugaeff and Zerevitinoff.—Ber., 40, 2027, 1907.
6 Shaffer and Hartman.—J. Biol. Chem., 45, 365, 1921.
7 Jacobs and Craig.—*Ibid.*, 119, 145, 1937.
8 Vesterberg.—Ber., 36, 4200, 1903.

IX. BLOOD GROUPING INVESTIGATIONS IN INDIA, WITH SPECIAL REFERENCE TO SANTAL PERGANAS, BIHAR.

By SASANKA SEKHER SARKAR.

(*Received 15th August, 1938.*)

Although blood grouping investigations have made rapid strides in all countries in recent years India is still lagging behind. Up till now only members of 43 castes and tribes have been grouped and even among some of them the percentages were calculated on scanty data. In a large number of cases the samples have been drawn from provinces as a whole, irrespective of caste or tribe. Dr. Guha [1] has rightly pointed out the value of such heterogeneous samples as negligible from the anthropological standpoint. Dr. Hutton [2] in his Census Reports, 1931, also called attention to the importance of blood group investigations on caste basis. Caste endogamy is an age-old institution in India and some of the upper castes have already acquired a selective value of much genetic importance.

Snyder [3] in the concluding paragraph of his book has stressed that 'the greatest need at present is the study of real races rather than nationalities or political groups'. The desideratum about 'real races' is not necessarily fulfilled by separating the individuals of each aboriginal tribe, but when we group together a number of aboriginal tribes we make a bigger muddle than by grouping a number of castes. Cases of inbreeding in small tribes or groups like the Paniyans [4] of Wynaad plateau and the white Jews [5] of Cochin are likely to affect the blood group percentages. Race crossing has taken place in varying degrees among almost all the castes and tribes and though a 'real race' can hardly be found, blood grouping investigations will no doubt throw light on the amount of inter-mixture a so-called race or tribe has undergone. Further, India has been suggested to be the land of origin of B mutation and as a result of the rare frequency of human mutations, as

shown by Gates [6] and Fisher, an extensive search is necessary to trace, if possible, the locus of the origin of B mutation.

The Data.

The present data were mostly collected from the district of Santal Perganas in Bihar. Some Bagdis from the district of 24 Perganas and Oraons of Chota Nagpur were also examined for purposes of comparison. The Oraons immigrated in the suburbs of Calcutta as field or day labourers.

Agglutination tests were made by the open slide method of Wiener with test sera of groups A and B supplied by the Haffkine Institute, Bombay. The tests were carried out during the winter months of 1937 and 1938. Frequent checks were made by repeating the tests and in a number of cases the samples were either retested by Dr. Macfarlane or jointly done by both of us. In the majority of cases washed red blood corpuscles were tested but in cases of emergency the whole blood had to be tested. Within a day of the arrival of the test sera, the tests were carried out in the field and work was finished within two weeks of the supply. The weekly markets were chosen to be the best spots for collecting blood samples. It was a matter of great difficulty to get the aborigines to consent to the pricking of their fingers, and the officers of the forest department in *dāmin* areas and the police officers in non-*dāmin* areas were of immense help to the author. The widespread abhorrence to the letting of blood is not only confined to the aborigines but to many backward classes of the Hindu community. Among the Santals no individual belonging to '*sādā*' sub-clans of a clan will allow himself to be pricked and I have not succeeded in doing so. The data collected are shown in Table I below :—

TABLE I.

Blood Groups from Bengal and Bihar.

Subjects.	Total	O	A	B	AB	p	q	r
Bengal :—								
Bāgdi ..	80	25	18	28	9			
	%	31·25	22·50	35·00	11·25	17·98	26·07	55·95
Māl ..	9	3	..	2	4			
Bihar :—								
Santāl ..	339	112	71	118	38			
	%	33·04	20·94	34·81	11·21	17·63	26·53	57·45
Hill Mālér ..	235	99	60	63	13			
	%	42·13	25·53	26·81	5·53	16·97	17·74	64·91
Plains Mālér	34	10	3	17	4			
	%	29·41	8·82	50·00	11·76	10·89	38·01	54·23
Mālpāhāriās	23	9	7	5	2			
	%	39·13	30·43	21·74	8·70	21·98	16·60	62·55
Orāons ..	35	9	9	11	6			
	%	25·71	25·71	31·43	17·14	22·72	26·56	50·69
Kumhār ..	26	10	8	4	4			
	%	38·46	30·76	15·38	15·38	26·62	16·80	62·02
Moslems ..	29	10	8	9	2			
	%	34·48	27·59	31·03	6·89	19·06	21·21	58·71

The other castes from Santal Pergs., Bihar, include the following:—

Teli (12), O8, Al, B3; Bhuiyā (10), O3, A3, B2, AB2; Bāuri (12), O4, A1, B6, AB1; Modak (11), O5, A2, B3, AB1; Mirdhā (7), O2, A2, B2, AB1; Mahāli (7), O4, A1, B2; Hāri (6), O2, A3, B1; Turi (6), O1, A2, B2, AB1; Āhir (7), O5, B2; Māl (4), O3, A1; Mochi O1; Kheturi O1; Nāpit (3), O2, B1; Rāoniār (2), O1, B1; Hālwai O1; Tāmbuli, O1; Bāniā B1; Mārayā (3), O1, A1, B1; Sonār, B1; Chattri, B1, Kāmār, (3) O2, A1; Kālwar, B1; Rājwar, O1; Kurmi, O1; Kāhār, O2; Chāsā, B2; Jāliā, B1; Suri, (5), O3, B2; Jugi (3), O2, B1; Bairāgi (2), A1, B1; Tānti (3), O1, B2; Brāhman (4), O1, A2, B1.

For the purpose of treating the data of the other groups they have been divided into the following five divisions:—

(I) The Māls have been grouped with the Mālpāhāriās because of their very close physical and cultural affinities. A very small sample of Māl from Bengal has also been tested.

(II) The other aboriginal tribes, Mahāli (7), Mārayā (3) and Bhuiyā (10) have been put together. Both the Mahāli and the Mārayā have very close affinities with the Santals and appear to have branched off as small occupational groups from the parent stock. The Mahālis earn their livelihood by basket-making while the Mārayās cast ornaments by smelting scrap metals.

(III) Some of the Hindu castes of very low social order, whose aboriginal heritage is beyond doubt have been grouped under one head:—

Hāri (6), Bāuri (12), Turi (6), Dom (7), Mochi (1) .. 32

(IV) The following Bengali speaking occupational castes have been grouped together; e.g. Bairāgi (2), Jugi (3), Modak (11), Tānti (3), Suri (5) and Tāmbuli (1) ..... 25.

(V) The Hindi speaking occupational castes mentioned below have been similarly classed together. They consist of Hālwai (1), Bāniā (1), Teli (12), Kālwar (1), Rājwar (1), Kāhār (2), Kāmār (3), Sonār (1), Rāoniār (2), Nāpit (3), Chāsā (2), Ahir (7), Kurmi (1) 37

The distribution of the blood groups of the above five groups are as follows:—

TABLE II.

(Blood Groups of other Castes, Santal Pergs.)

	Total	O	A	B	AB	p	q	r
Group I ..	27	12	8	5	2			
	%	44·44	29·63	18·52	7·41	20·65	12·94	66·66
Group II ..	20	8	5	5	2			
	%	40·0	25·0	25·0	10·0	19·38	19·38	63·25
Group III ..	32	10	8	11	3			
	%	31·25	25·0	34·38	9·38	18·99	25·0	55·90
Group IV ..	25	12	3	9	1			
	%	48·0	12·0	36·0	4·0	20·0	22·54	69·28
Group V ..	37	23	2	12	..			
	%	62·16	5·41	32·43	..	2·74	17·79	78·84

Analysis of the Data.

The hill Mālérs [Plate 11, Figs. 1 and 1(*a*); 2 and 2(*a*)] are the most primitive people in the district of Santal Perganas. They live on the hill slopes which are the most inhospitable areas of the district. Agriculture is very crudely practised. Maize is grown to a large extent and unlike the people of the plains, maize forms their staple food. The physical characters [7] and the social life [8] of these people have been described by the author in earlier publications.

The plains Mālérs [Plate 12, Figs. 3 and 3(*a*)] have now begun to settle on the plains and their number is very few. This appears to be due to purely economic reasons as the forests are gradually disappearing and food is getting scarcer every day. The plains Mālérs are now trying to obtain lands on the plains and in fact, a few have already acquired some.

The Mālpāhāriās [9] branched off from the Mālérs sometime during the 18th century. There are very strong evidences to show that originally the Mālpāhāriās are culturally, physically and linguistically, a part and parcel of the Mālérs. In physical features neither of the above three show any wide divergence.

The blood groups of the above three peoples also agree with one another, but they show varying degrees of admixture. The hill Mālérs have the highest percentage of O (42·13%) while the percentages of A (25·53) and B (26·81) are almost equal. The percentage of A approaches that of some South Indian tribes and castes (see Tables VI and VII). The plains Mālérs on the other hand show a very high percentage of B (50·0%) and a very low A (8·82%). The blood groups of the plains Mālérs were collected mostly from Godda Subdivision (Rajabhita, 17; Simra, 12) where B is predominant among the Santals also (Table I).

The Mālpāhāriās, like the hill Mālérs, disclose the highest percentage of O (39·13%), but the percentage of A (30·43%) is higher than that of the hill Mālérs. The number of the Mālpāhāriās is, however, too small to warrant any definite

The other five plains Mālérs were from Berhait, Rajmahal.

conclusion though this feature is characteristic of the hill Mālérs also.

In the preliminary note [10] published in 'Current Science' by the author the percentages of the hill Mālérs given there are not similar to those published in Table III.

TABLE III.

	O	A	B	AB
Hill Mālér	65	22	44	8
139	46·76%	15·83%	31·65%	5·76%
Hill Mālér	99	60	63	13
235	42·13%	25·53%	26·81%	5·53%
Hill Mālér	34	38	19	5
96	35·62%	39·58%	19·79%	5·21%
Santal	64	41	70	24
199	32·16%	20·60%	35·18%	12·06%
Santal	112	71	118	38
339	33·04%	20·94%	34·81%	11·21%
Santal	48	30	48	41
140	34·29%	21·43%	34·29%	10·0%

It will be evident from the above table that while the three series of data of the Santals agree closely with one another, those of the hill Mālérs show a wide divergence in respect of the percentages of the blood groups A and B. The data collected from the villages situated on the bank of the River Gumāni in February 1938 is highly interesting in having the highest percentage of A (39·58%) and the lowest of B (19·79%); this may not improbably be due to high inbreeding within this locality. The high percentage of A in this locality will be further evident from the distribution of the blood groups in the various localities, as shown in Table IV.

A high preponderance of the agglutinogen A was noticed by the author in the early parts of the tour in 1937 when he was working at Kusma, a village situated on the bank of the River Gumāni. Kusma has also a large number of Kumhārs (potters) and as will be seen from the blood groups percentages given in Table I the high percentage of A (30·76%) is also present among them. The Kumhār sample is however too small in comparison with that of the hill Mālérs.

TABLE IV.

Blood Group distribution according to localities (Bungalows).

Bungalows.	Subdivision.	Hill Mālérs.				Santals.			
		O	A	B	AB	O	A	B	AB
Brindabani ..	Rajmahal	20	8	10	..	20	8	14	5
Borio ..	,,	11	13	15	2	6	8	5	3
Berhait (Gumāni) .. Plains	,,	1	..	2	2	..	..	..	..
Hill	,,	6	3	4	1	8	7	15	13
Taljhari ..	,,	3	1	2	..	1	..	1	..
Madro ..	,,	9	..	8	..	6	..	2	..
Durgapur (Gumāni)	,,	6	5	2	1	10	5	9	1
Raxi ..	,,	..	..	1	..	1	..	..	2
Kusma (Gumāni) ..	,,	13	5	4	2	9	5	6	3
Banjhi ..	,,	4	2	1	2	3	6	14	2
Maharajpur ..	,,	4	1	5	..	4	2	6	..
Ranga (Gumāni) ..	,,	..	..	..	..	..	3	4	..
Sakrogarh ..	,,	10	2	2	2	3	..	2	1
Patna (Gumāni) ..	,,	3	..	..	..	..	..	..	..
Parerkola ..	Pakur	..	..	..	..	..	..	1	1
Simlong (Gumāni) ..	,,	2	1	3	..	..	1	1	..
Litipara ..	,,	..	..	..	..	..	1	..	..
Dharampur (Gumāni)	,,	2	5	..	1	7	3	4	1
Hiranpur ..	,,	..	..	..	..	10	4	6	5
Surma ..	,,	1	5	..	..	..	..	..	..
Dhamni (Gumāni) ..	Godda	5	7	2	2	5	4	6	..
Rajabhita Plains ..	,,	6	2	7	2	1	1	5	1
Simra ,, ..	,,	3	1	8	..	4	1	3	..
Boarijore ..	,,	..	..	3	..	3	5	6	..
Karmataur ..	,,	..	1	..	..	..	..	..	..
Chandna ..	,,	..	..	1	..	..	..	..	..
Gando ..	Dumka	..	..	..	..	10	7	7	..

The Kumhārs are clay hunters and their occupation necessitates living on the river banks. The River Gumāni is the largest river of the district and possesses the largest valley. It has attracted the earliest settlement of people and even at present forms the most populous and the most fertile land in the district. It would be interesting to compare the somatic characters of these two groups and find out whether they originally formed part of the same stock or not.

The hill Mālérs are also very strong in O. The high percentage of A is largely prevalent among the South Indian

tribes, as has also been noticed among some of the Mongoloid tribes of N.E. India. The geographical position of the hill Mālérs in the eastern part of the Central Indian plateau makes it equally possible for them to have derived this characteristic gene A from either of the two sources. The hill Mālérs however, have a high percentage of B which is found among the non-aboriginal groups of South India. The Mālpāhāriās also agree with the hill Mālérs in having the highest percentage of O and a high percentage of A. Thus the physical and cultural affinities of the two, as shown by the writer, appear to receive further corroboration from the serological data. The dual origin of the Mālérs and the Mālpāhāriās, as advocated in the last census of 1931, therefore do not seem to be based upon much scientific evidence.

The Santals [Plate 12, Figs. 4 and 4(*a*) and Plate 13, Fig. 5] have the highest percentage of B (34·81%) followed closely by the Orāons (31·43%). The latter, however, have a higher percentage of A (25·71%) than the Santals (20·94%). Both the Santals and the Orāons (Plate 13, Fig. 6) agree with the Bāgdis of Bengal in the blood groups percentages.

The blood groups percentages of the other castes of Santal Perganas, divided into five groups, are given in Table II. There is a clear line of demarcation between the aborigines and the semi-aborigines, who show high percentages of both the agglutinogens A and B, while the Hindu castes only predominate in the agglutinogen B. The non-agglutinogenic factor O is found in the highest percentages among the upper caste Hindus. The Moslems also agree with aborigines and semi-aborigines in their blood group compositions.

The Bāgdis[12] [Plate 13, Figs. 7 and 7(*a*)] of Bengal occupy the same social status as group III of Santal Perganas. In fact their physical characters closely link them up with the Santals of the Santal Perganas, where they are known as Bāuris and live side by side with the Santals. The 12 Bāuris tested serologically are as follows:—

O	A	B	AB	Total
4	1	6	1	12

Compared with the aborigines of Santal Perganas they show the following values:—

TABLE V.

	O	A	B	AB
Bāgdi (Bengal) ..	25	18	28	9
%	31·25	22·50	35·50	11·25
Group III	10	8	4	3
%	31·35	25·0	34·38	9·38
Santal	112	71	118	38
%	33·04	20·94	34·81	11·21

The close resemblance between the Santals and Bagdis, also pointed out by Dr. Macfarlane,[12] is thus very apparent.

Indian Blood Group Position.

A glance at the map (Plate 14, Fig. 8) will show the scanty blood group data from India. Nothing is known of a large part of Central and Western India. Thanks to the efforts of Da Silva Correia, we have some data from Nova Goa. In Southern India the largest data is from Cochin, due to the efforts of Dr. E. W. E. Macfarlane, who is also responsible for a large collection of data from Bengal. Capt. Mitra is responsible for a large number of data from Assam while Malone and Lahiri have supplied mass data from Northern India. In the absence of any data from the regions indicated in the map no generalization is possible though some well-defined zones of blood group concentration are clearly marked.

In Tables VI and VII all the available blood group results have been arranged and classified firstly, according to their common anthropological name and secondly according to geographical areas.

TABLE VI.

Mongoloids.

	O	A	B	AB	
Tibetans	46·5	35·7	12·5	5·3	Macfarlane.[13]
Tibetans	14·9	35·7	13·9	24·1	Tennants.[14]
Nepalis	33·3	34·6	23·1	9·0	Macfarlane.[13]
Lepchas	24·0	36·0	32·0	8·0	„
Angami Nagā ..	46·06	38·78	11·52	3·64	Mitra.[15]
Lushai	32·63	44·68	16·31	6·38	„

Aboriginal Tribes.

(South India and Eastern India.)

		O	A	B	AB	
South India	Illuvas ..	58·3	24·2	12·2	5·3	Macfarlane.[16]
	Pre-dravidian Tribes (Madras) ..	48·0	30·0	9·0	12·0	,,
	Paniyans ..	20·0	62·4	7·6	10·0	Aiyappan.[4]
	Todās ..	29·5	19·5	38·0	13·0	Pandit.[18]
Eastern India	Santal ..	33·04	20·94	34·81	11·21	Sarkar.
	Mālé ..	42·13	25·53	26·81	5·53	,,
	Plains Mālé ..	29·41	8·82	50·00	11·76	,,
	Mālpāhāriās ..	39·13	30·43	21·74	8·70	,,
	Orāons ..	25·71	25·71	31·43	17·14	,,
	Dravidian Tribes ..	24·3	27·5	36·8	11·4	Malone and Lahiri.[17]

The following will be evident from Table VI:—

(1) The Mongoloid peoples living on the north-eastern frontier of India have among all of them a fairly high percentage of A. Among these the highest percentage of B are found among the Lepchas (32·1 %) and the Nepalis (23·1 %). Both the latter data were however collected by Dr. Macfarlane [13] from Kalimpong, Bengal.

(2) The aboriginal tribes of South India possess a high percentage of A while that of B is very small. The aborigines occupying the eastern portion of the Central Indian highlands show the presence of an almost equal percentages of the two agglutinogens. Both the two groups, however, agree in having a high frequency of A.

(3) The Todās [18] of the Nilgiri Hills stand aloof from other South Indian tribes in having the highest percentage of B.

TABLE VII.

South India.

	O	A	B	AB	
Nairs ..	38·8	35·5	22·4	3·3	Macfarlane.[16]
Syrian Christians	36·4	26·4	28·6	8·6	,,
Tamil Non ..	42·0	28·0	24·0	6·0	,,
White Jews Cochin	18·0	62·0	20·0	..	,,
Black Jews ,,	73·6	10·4	16·0	..	,,
Tamil coolies ..	37·9	23·0	31·6	7·5	Bais and Verhoef.[21]

Eastern India.

	O	A	B	AB	
Bengali Kayasthas	38·9	20·8	32·5	7·8	Chaudhuri.[19]
Bengali (mixed) ..	37·9	19·3	34·1	8·7	,,
Kumhar (Santal Pergs.) ..	38·46	30·76	15·38	15·38	Sarkar.
Muslims ,, ..	34·48	27·59	31·03	6·89	,,
Mahishya (Beng.)	32·5	20·0	39·4	8·1	Macfarlane } unpub-
Pods ..	31·11	17·78	44·44	6·67	,, } lished.
Beng. Muslims ..	28·3	23·3	40·0	8·3	,, }
Bāgdis ..	31·25	22·50	35·0	11·25	Sarkar & Macfarlane.
Assamese ..	33·65	24·55	32·55	9·25	Mitra.[20]
Santal Pergs. low castes ..	47·14	14·05	33·88	4·96	Sarkar.

Northern India.

	O	A	B	AB	
Baluch ..	47·2	24·3	24·3	4·2	Malone & Lahiri.[17]
Hazaras ..	32·0	25·0	39·0	4·0	,,
Pathans ..	29·3	31·3	33·3	6·1	,,
Jats ..	33·2	24·5	35·5	6·8	,,
Rajputs ..	28·8	28·0	33·0	10·2	,,
Khatris ..	33·3	25·3	30·3	11·1	,,
U.P. Hindus ..	30·2	24·5	35·5	6·8	,,

Western India.

	O	A	B	AB	
Hindus (Goa) ..	33·5	24·5	31·0	11·0	de Figueiredo.[21]
Maharathas (Goa)	29·25	26·75	34·0	10·0	Correia.[21]
Indian Christians (Goa) ..	31·6	22·65	30·42	15·85	de Figueiredo.[21]

The position of the other non-aboriginal groups can be summarized as follows:—

(4) The various South Indian castes (five of which are however from Cochin) show the presence of B in various percentages from 16·0% to 31·6%. Like the aborigines of eastern Central Indian highlands some possess an almost equal amount of the two agglutinogens A and B.

(5) The blood group positions of Eastern India (excepting the frontiers of Assam), Northern India and Western India are

similar. All are characterized by the presence of a high frequency of B while that of A varies from 14·05% to 28·0%. The Kumhārs of Santal Perganas and the Pathans of North-Western Frontier however, show a higher frequency of A. With the exception of the plains Mālérs, who appear to be highly intermixed, the highest percentage of B is recorded from Bengal among the Pods (44·44%) an agricultural caste of Bengal.

The position of Bengal in respect of the frequency of the blood Group B is interesting. The province shows the presence of the highest percentage of B among some of the lower castes (Table VII) and to the north and north-east of Bengal we have correspondingly strong centres of A group concentration. Can the locus of origin of B mutation be, therefore traced here?

In conclusion the author acknowledges his best thanks to Dr. E. W. E. Macfarlane, Ph.D., D.Sc., for her constant help in course of this work and also for her allowing the author to use her unpublished data of Bengal blood groups. The author also expresses his thanks to Dr. B. S. Guha, M.A., Ph.D., Anthropologist, Zoological Survey of India, for his valuable suggestions.

BIBLIOGRAPHY.

[1] Guha, B. S.—Progress of Anthropology during the past twenty-five years, p. 323, Calcutta, 1938.

[2] Hutton, J. H.—Census of India, 1931, Vol. I, Pt. I, p. 451, Delhi, 1933.

[3] Snyder, L. H.—Blood Grouping in relation to clinical and legal medicine, p. 135, Baltimore, 1929.

[4] Aiyappan, A.—Blood Groups of the Paniyans of the Wynaad Plateau, Man, 1936, 255.

[5] Macfarlane, E. W. E.—The racial affinities of the Cochin Jews, Jr. Roy. Asiatic Soc. Bengal, Letters, Vol. III, 1937, No. I, p. 15.

[6] Gates, R. R.—Blood Groups and Physiognomy of the British Columbian coastal Indians, Jr. Roy. Anth. Inst., LXIV, 1934, p. 32.

[7] Sarkar, S. S.—The Racial Affinities of the Inhabitants of the Rajmahal Hills, Trans. Bose Res. Inst., XII, 1936-37, p. 123.

[8] Sarkar, S. S.—The Mālers of the Rajmahal Hills, Calcutta, 1938.

[9] Sarkar, S. S.—The Origin of the Mālpāhāriās, The Indian Hist. Quarterly, IX, 1933, p. 886.

[10] Sarkar, S. S.—Preliminary Note of the blood groups of the Aborigines of Bihar, Current Science, VI, 1937, No. 6, p. 283.

[11] Ghosh, R.—Note on the Sauria or Mãler Paharias, Census of India, Vol. I, Pt. III, p. 112, Delhi, 1935.

[12] Macfarlane, E. W. E.—Supplementary Note on the blood groups of the aborigines of Bihar, Current Science, VI, 1937, No. 6, p. 284.

[13] Macfarlane, E. W. E.—Eastern Himalayan Blood Groups, Man, 159, 1937.

[14] Gates, R. R.—Tibetan Blood Groups, Man, 147, 1936.

[15] Mitra, P. N.—Blood Groups of the Angami Nagã and the Lushei Tribes, Ind. Jr. Med. Res., XXIII, 1935-36, p. 685.

[16] Macfarlane, E. W. E.—Preliminary note on the blood groups of some Cochin Castes and Tribes, Current Science, V, 1936, p. 653.

[17] Malone, R. H. and Lahiri, M. N.—The distribution of the blood groups in certain races and castes of India, Ind. Jr. Med. Res., XVI, 1928-29, p. 963.

[18] Pandit, S. R.—Blood Group distribution in the Todas, Ind. Jr. Med. Res., XXI, 1933-34, p. 614.

[19] Chaudhuri, A.—Preliminaries to a study of the racial problem in India, Man in India, XVI, 1936, p. 16.

[20] Mitra, P. N.—Blood Groups and Heredity, Ind. Jr. Med. Res., XXII, 1934-35, p. 495.

[21] Correia, Da Silva, A. C. G.—The Blood Groups in Portuguese India, Proc. Ind. Academy of Sciences, B, Vol. III, 1936, p. 115.

FIG. 1.

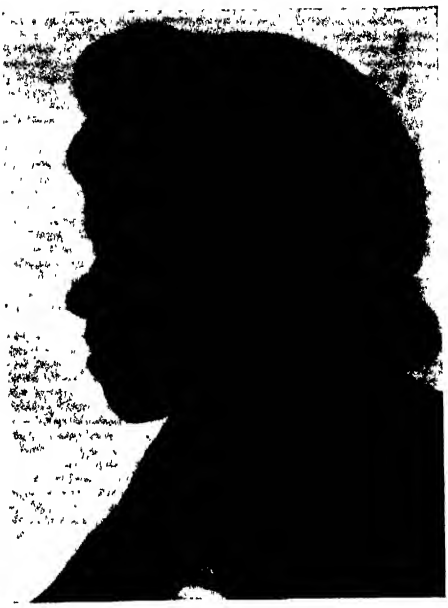


FIG. 1(*a*).

Hill Mālér, Maharajpur, Blood Group, O.

FIG. 2.

FIG. 2(*a*).

Hill Mālér, Berhait, Blood Group, O.

FIG. 3.

FIG. 3(*a*).

Plains Mālér, Rajabhita, Blood Group, B.

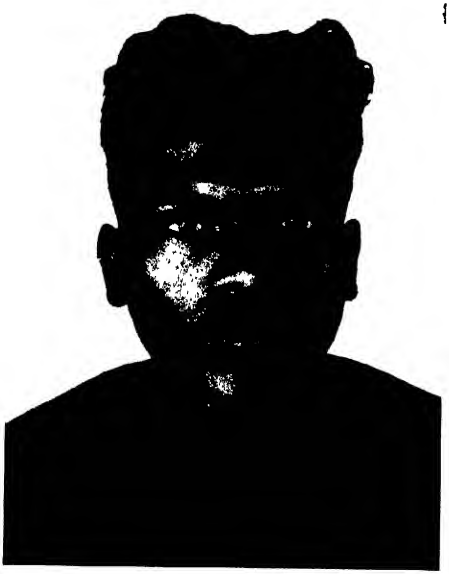


FIG. 4.

FIG. 4(*a*).

Santal, Simra, Blood Group, O.

FIG. 5. Santal, Brindabani,
Blood Group, B.

FIG. 6. Oraon mother with child,
Blood Group of both, AB.

FIG. 7.

FIG. 7(*a*).

Bagdi, Bengal (Diamond Harbour) Blood Group, AB.

MAP OF INDIA

showing

Distributions of the Blood Groups A & B.

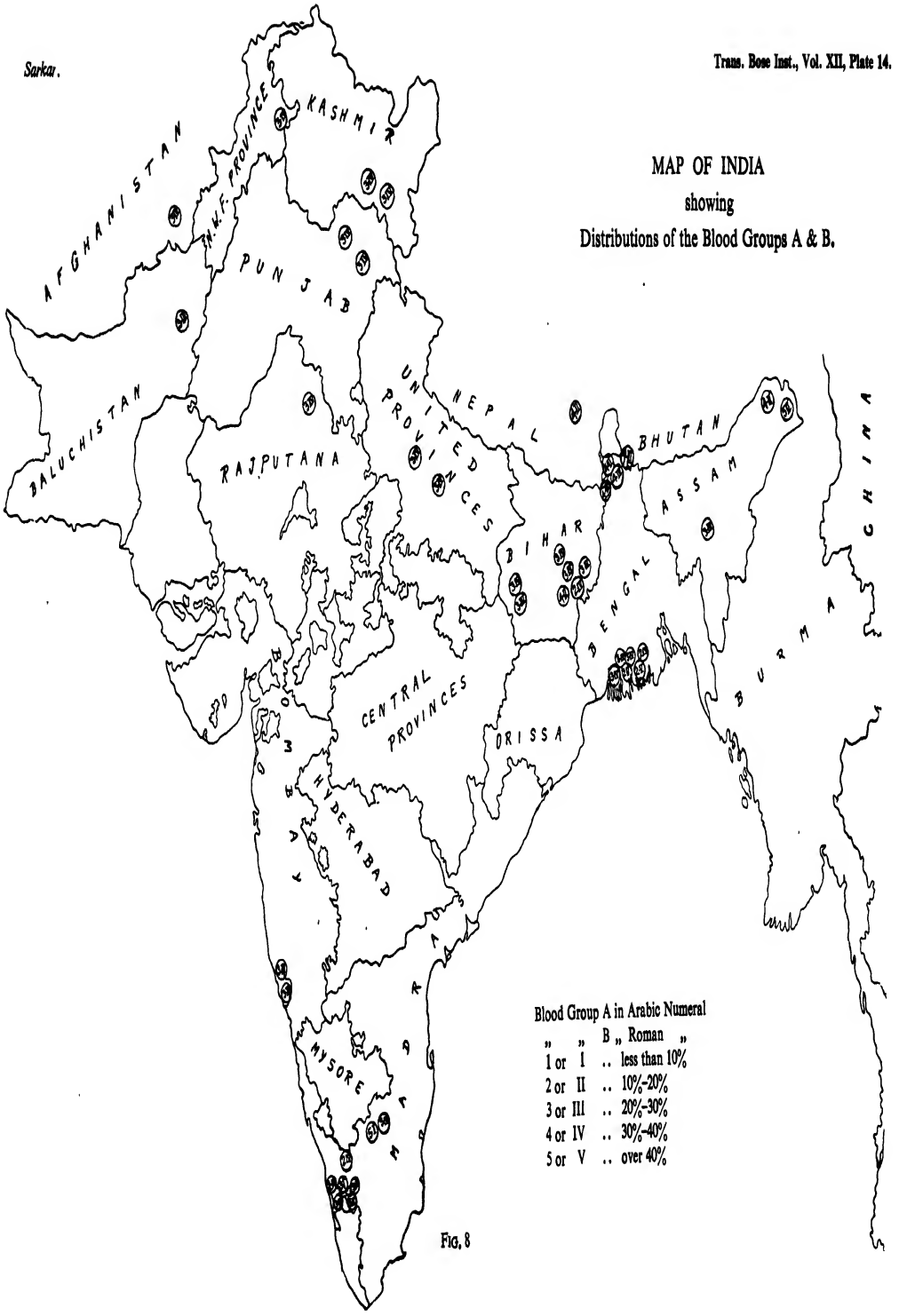


Fig. 8

X. ON THE MOULTING AND METAMORPHOSIS OF *MYRMARACHNE PLATALEOIDES*, CAMB.

By GOPAL CHANDRA BHATTACHARYA.

(*Received 15th August, 1938.*)

INTRODUCTION.

The general process and different aspects of moulting in different spiders have been studied by eminent Arachnologists,[1–16] but the moulting process of the spiders of the ant-mimicking group, particularly that of the common Indian species *Myrmarachne plataleoides*, Camb., has not, so far as I am aware, been described. A knowledge of the form at each stage is of great value in determining specific identity, as for lack of it there is a danger that spiders of the same species at different stages of their development may be described as the distinct and different species. Pickard Cambridge [17] and the Peckhams [18] who dwelt at length on the systematic study of these spiders do not seem to have paid attention to the details of their moulting process. The subject of this paper is to give a detailed description of the moulting process of the ant-mimicking spider, *M. plataleoides*, Camb., with special references to the male forms of the species, and to show that sexual dimorphism appears during the final moult.

Sometime ago I happened to come across an apparently female specimen of *M. plataleoides*, Camb., lying under its silky retreat on a leaf of *Calophyllum inophyllum*, in the Royal Botanical Gardens at Sibpore, near Calcutta. I collected the specimen and kept it in a glass tube and the following morning I was surprised to note a remarkable change in the contour of the body. This specimen had by now become a fully developed male, provided with the copulatory organs at the digital joints of the palpi, the chelicerae being nearly half as long as the body. These characteristics, as already stated, were not at all noticeable on the previous day when the specimen was

collected. This marked transformation led me to investigate the details of the moulting habits of this species, particularly with reference to the changes that occur during the final moult.

Method of Rearing.

From among a number of live specimens of both sexes of *M. plataleoides*, Camb., I selected a well developed pair, the female, after mating, being kept in a separate tube and observations on it recorded in the laboratory. The spider was fed on a diet consisting of minute winged insects, milk and water. Spiders and insects always move vertically up when they are confined in narrow tubes or in narrow elongated vessels. I took advantage of this habit in introducing living insects as food and in transferring spiders from one tube into another without handling them. The spider in the tube was examined under a magnifying lens.

Method of Photographing.

Some clean glass plates were thinly coated with a dilute solution of fish glue in water and then dried. The transparency of the plate was hardly affected by this treatment. By pasting a white or black paper on the reverse face of the plate a suitable background was obtained. The plates were then serially numbered with glass pencils. Each of the spiders, which were reared in separate tubes, numbered serially, was then allowed to escape from the tube by removing its cotton plug, in such a way as to make the spider come in contact with water, contained in a shallow vessel and thus get wet. It was then allowed to leave the water and course back along the drag-line attached to the tube. The wet spider was brought in contact with the specially prepared plate already described. The creature crawled over the plate and soon got stuck on the glued plate which became hardened within a few seconds after the evaporation of the moisture. The spider, exhausted by its constant struggle to free itself from the adhesive film, lay down motionless. The spider thus became fixed to the plate. The legs and parts of the body were then properly stretched by manipulation. The glass plate with the live specimen fixed to it was then placed before

the camera and snap-shots taken. After the photographs had been taken, a drop of water was poured over the spider so as to dissolve away the adhering glue. A hair brush was placed before the spider which it readily caught hold of and crawled up. Later it was carefully transferred to an observation tube. Series of photographs were taken of each spider at different stages of its life, without interfering in any way with its moulting process and life conditions. After the photographs were taken each of the spiders was reared till maturity was reached. As it was not possible to determine the sex of these immature specimens at the time of photographing, I had to wait till the final moulting for the determination of sex.

OBSERVATIONS.

The mother-spider lays at a time 8 to 12 globular eggs, light yellow in colour, under its retreat made of fine spun silk. In the course of 12 to 15 days the young spiders come out and remain under the retreat without taking any food for about 5 to 6 days. The young spiders at this stage are yellow in colour with a continuous black line over the anterior row of eyes. After the lapse of 5 or 6 days the young ones emerge from the retreat. These young spiders measure about 1 to 1·5 mm. in length. At this stage, the dorsum of the cephalothorax is almost black with a slight yellowish tint. The anterior portion of the abdomen, both dorsally and ventrally, is orange or reddish yellow in colour, while the posterior half is deep black. The eyes are more protruding and placed closer to each other than those in the adult stage. The cephalic portion is high and flat, but the thoracic portion slopes with a slight upward curvature in the middle; the abdomen is oval with a slight depression on the dorsum.

In the course of 7 to 10 days after emergence from the retreat, the young spiders change their skins. This I have taken as the first moult. The first moulting may sometimes take a longer period, depending upon various factors. The second moult then follows 10 to 15 days after the first, a slight depression appearing at the middle of the cephalothorax and the abdomen.

The young spider now appears rather broad than long, owing to the shortness of the pedicle. But after the third ecdysis, the body becomes elongated and assumes the form characteristic of the species. The blackish shade of the anterior portion of the body acquired after emergence of the young from the cocoon is by this time replaced by a reddish yellow tint while the region round the eyes retains the original blackish shade that persists throughout life. In rare instances, however, the young ones after leaving the cocoon are found absolutely black and the abdominal colour variations appear only after the third ecdysis. Two or three days after the fourth ecdysis, the abdominal black tint disappears within a few hours. An orange or yellow colour then becomes prominent all over the body with a few black specks on the cephalic region. The spider now measures approximately 3 to 5 mm. in length. White markings, characteristic of the adult stage, now begin to appear on the abdomen and on the sides of the cephalothorax. It is worth noting that in this species, *M. plataleoides*, the appearance or disappearance of markings or spots follows each fresh moult. This is in agreement with what has been observed by Pollock [19] in the moulting of *Epeira aurelia* where the spots on the sides of the abdomen gradually disappear giving rise to very handsome markings. After the fifth moult the spider approaches the normal size of an adult, the colour becoming brick-red with distinct white markings on the dorsal aspect of the abdomen and on the sides of the cephalothorax. Immediately before the sixth moult the average length of the spider is about 8 to 10 mm.

It will be of interest that these spiders at different stages of development mimic different species of ants. The mimicking habit of them had been observed by different authors in the mature specimens only. [20–24] I have noticed *M. plataleoides* at their first, second and third stages to mimic *Solenopsis geminata*, and after the fourth moult the ants, *Plagiolepis longipes*, Jerd. After the fifth moult the full grown adult female perfectly mimics *Œcophylla smaragdina*, Fabr. In these respects my observations slightly differ from those of Dr. Mathew. [25]

Uptil the final (i.e. the sixth) moult all the immature forms resemble mature females, the sexes becoming distinct after the

final moult. The habit, biology, external structures, size, colour and markings of the body and even the number and arrangement of teeth in the chelicerae of the sex-undetermined immature forms are in all respects similar to those of the mature females. [Fig. 1 (*a*), (*b*), Plate 15.]

The tibial and tarsal joints of the pedipalpi of both mature and immature forms are identical, being oval and flat in shape. Two deep rounded depressions on the epigyne are present in both the cases. The only noticeable difference is that the depressions in the mature female are more definite than those in the immature forms. The distinction between the two sexes at earlier stages, namely before the final moult in this species of spiders is so negligible that immature specimens cannot be separated according to their potential sexes. In other species of spiders, however, the sex can generally be easily distinguished even at earlier stages by comparing the digital joints of the pedipalpi of the two sexes.

From laboratory observations it appears that even an adult male spider fails to distinguish between a sex-undetermined immature form and a fully developed mature female. I have often observed male spiders performing the various amorous acts, prior to mating, before an apparently female specimen (i.e. immature specimen before the final moult). The male spider tries to allure the immature specimen by caressing with its club-like chelicerae and front pair of legs. Mating, however, does not take place in this condition inspite of their innate sexual difference. It may not be out of place to mention in this connection that these spiders do not allure the females by dancing as has been observed in other spiders.[26]

I shall now turn to the changes that take place after the sixth or the final moult. The armature of the epigynum characteristic of the adult female now becomes fully developed, and the colour of the atriolum becomes dark brown and conspicuous. The immature forms of the female sex by this time become sexually mature. Mating does not take place until this stage is reached. But Pollock [27] says of *E. aurelia* that 'about a week after the fifth cocoon has been made, the spider changes its skin for the last time, rests its egg-laying for about thirty days,

makes five more cocoons at intervals of from 16 to 25 days, and dies a week or so after making its last one.'

Very striking changes take place in the immature forms of male sex (potential) during the final moult. The last transformation into a sexually mature male form takes place within about half an hour.

Two or three days previous to casting off its last integument, the immature spider spins an oval-shaped simple retreat composed of loosely woven silk and rests within it without taking any food. The spider has been observed not to move out of the retreat of its own accord. If, however, it is induced to prey upon some insects and suck the juice of the victim, the moulting is observed to be delayed for a considerable period of time. During the progress of moulting this species does not attach itself to the walls of the retreat by means of threads emitted from the spinnerets, but attaches itself to the retreat by the claws of its legs. This attachment by means of claws is of vital importance to the spider in its moulting. The old chitinous cuticula covering the dorsum of the cephalothorax is the first to give way along the sides and is raised up like a lid, its posterior extremity remaining fixed to the abdominal slough. [Fig. 2 (*a*), Plate 15.] The body then works itself out and the appendages are gradually disengaged from their exoskeleton. During this progress the chelicera is drawn out from its old cuticle and undergoes growth and transformation while still soft. The pedipalpi are then drawn out of the slough along with first and second pair of legs. The sex of the spider becomes evident no sooner than the palpi have come out of the old cuticle before the abdominal skin has finally been cast off. The palp, which was flat before the last moult, now becomes rounded and provided with a copulatory bulb and a tibial apophysis or copulatory claw, as we might call it, since it plays an important part during the act of mating. [Fig. 3(*b*), Plate 17.] Emerton [28] and others also have referred to a tibial hook of male Attidæ but have not described its function. Bristowe [29] mentions of a tibial spur of *Micrommata virescens*, Clerck, which is a straight thorn-like projection.

When the newly formed dorsal shield of the cephalothorax has become exposed by the uplift of the old cuticle, a pair of protuberances appear on the fresh surface of the cephalothorax at the site of the old small mandibles, which are often left behind with the old slough. These protuberances give rise to the chelicerae of the final form of the spider and require a detailed description.

These protuberances rapidly grow many times (about 15 to 20) longer and thicker than the original mandibles and project out along the long axis of the body. At first globular swellings with rudimentary uncinate processes appear at the apices of these protuberances. As growth takes place, these swellings gradually elongate giving the chelicerae the shape of Indian clubs with their upper and inner surfaces flat and fixed to the cephalothorax by the thin ends. Along with their growth the hook-like uncinate processes atriculated at the apices of the chelicerae also grow taking a backward turn along the under surface of the chelicerae and extend as far as their bases. [Fig. 3 (*d*), Plate 17.] The length of the uncinate process equals that of the chelicera and it is shaped like an elongated 'S'.

This whole process, that is from the appearance of the protuberances to the limit of growth, takes about 8 to 10 minutes' time. Two rows of teeth develop along the upper and lower margin of the inner flat sides of each chelicera. The upper margin which is known as the inferior ridge gets armed with about 12 to 14 teeth, all of which are bent forward and placed at equal intervals throughout the whole length, while the lower margin or the superior ridge contains five teeth equally distributed in the fore half of the chelicera. Sometimes the first two teeth near the tip of the chelicera, in the superior ridge, are fused together in one broad tooth. The teeth in the superior ridge are stronger than those in the inferior ridge.

The basal half of the chelicera, when viewed dorsally, looks transversely rugose and the extremity of it assumes abrupt enlargement, giving the spider much the appearance of spoon-bill, [Figs. 2 (*d*) and 3 (*d*), Plates 16 and 17]. At first these chelicerae look whitish but gradually take up a red tint. Black spots, which were absolutely absent before the moulting, appear at the apices

of the chelicerae. The third and fourth pair of legs come out of the old cuticle almost simultaneously along with the abdomen.

The whole process of the last moult is completed within half an hour or so. The spider sometimes experiences great difficulty in taking out the new mandibular appendages from its old cuticle. At times the first and second pair of legs are completely liberated before the extrication of the swelled anterior portion of the chelicera, resulting in the slough dangling about it. [Fig. 2 (*c*), Plate 16.] The spider can seldom shake off this slough and the failure to get rid of it ultimately causes its death. It has also been observed, at least in two cases, that when the grip of the retreat by the claws became detached either due to jerking or for other reasons at the time when the anterior half of the abdomen was being disengaged, the remaining portion of the abdomen could not come out. The spider tried to cast off the slough which hung from the abdomen and its failure to do so brought about its death.

After the moulting is over, the spider takes rest for a considerable period of time during which the whitish colour of the body turns brick-red. Dorsally, the abdomen of the male becomes highly constricted, while that of the female after the final moult is somewhat shallow and greatly distended. Sexual dimorphism is striking owing to the peculiar mandibular structure of the male. [Figs. 4 (*a*) and 4 (*b*), Plate 17.]

Discussion.

Blackwall [80] remarks that the process of moulting in different spiders is a uniform one. Although my experience is not in full agreement with this remark, it is, however, worth pointing out that while in *M. plataleoides* the sexes cannot be distinguished before final moult has been completed, there are other instances of existence of sexual dimorphism, particularly with regard to the development of the palpal organ, even before the penultimate moult. Spiders of the genera *Lycosa*, *Selenops*, etc., may be cited as examples in this connection. In some other cases, as in *Epeira*, *Nephila*, *Misumena*, etc., the sex can be differentiated long before the final moult by characteristic difference in markings, shape and colour of the body. Reference

may be made to the statements of other authors in support of the above observation. Comstock [31] observed that 'Previous to this last moult the tarsus of the pedipalp of the male is merely a club-like segment in appearance'. Wagner [32] says 'Pendant la periode des premieresumes, les males des Araignées ne se distinguent point des femelles, comme on le sait. Durant le developement ulterieur, les distinctions appraissent bien avant la dermiere mue.....' McCook's [33] view that 'These organs (club-like palpal organs) are almost devoid of soft part. It follows necessarily that as the shell cannot come off, no further moulting can take place' is not in agreement with the most common-place observation that both male and female spiders cast off their shell-like chitinous integuments of mandibles and fangs in each moult. While drawing attention to the difference between male and female spiders before maturity is reached Darwin [34] in his 'Descent of Man' says, 'I am informed by Mr. Blackwall that the sexes whilst young usually resemble each other and both of them undergo great changes in colour during their successive moults before arriving at maturity. In other cases the male appears to change colour. Thus the male of the above bright coloured *Sparassus* at first resembles the females and acquiring the peculiar tints only when nearly adult.'

This observation is important in view of Simon's statement [35] that only after the final moult the secondary sexual characters become apparent. Whilst this holds good in the present instance, viz. in *M. plataleoides*, such a generalisation, however, is hardly tenable in the other cases cited above.

Different authors [36–41] assert that spiders do not undergo any considerable change except in the development of palpal organs during the process of growth by moulting, but my experience with the species *M. plataleoides*, which undergoes altogether six moults, shows that an immature specimen (potential male) is totally changed at the final moult.

SUMMARY.

Though it is generally believed that the spiders do not undergo any metamorphosis except the palpal organs in the male at the final moult, yet a kind of transformation not only in

the tarsal segment of the pedipalp of the male but also in the chelicerae, teeth and fangs takes place in some species of Attid spiders, specially in *Myrmarachne plataleoides*, Camb.

Both the male and female of *M. plataleoides* change their skins generally for six times and after the sixth moult they become sexually mature and then no further ecdysis takes place.

Until the fourth ecdysis the colour pattern of the body, specially black on the posterior half of the abdomen, is most conspicuous.

During the sixth ecdysis, the immature specimen (potential male) is transformed into a distinctive male with its club-like chelicerae and copulatory organs with a tibial claw on each of the palpi. The arrangement of teeth is completely changed. Doubly bent fangs become as long as the stalk of the chelicerae. In the female, however, after the sixth moult, only the epigynum becomes conspicuous with two deep rounded depressions in the middle of the chitinous armature.

In many classes of spiders, the males and females differ in colour, size and shape by which the sex can be determined long before the maturity is reached. And in some cases the males can be recognised before maturity by the growing bulb at the tarsal segment of the pedipalpi. But in the case of *M. plataleoides* the immature forms of both the male and female cannot be differentiated until the final moult is reached. Even the sexually mature males cannot distinguish between mature females and sex-undetermined immature forms.

I gratefully acknowledge the generous help, constant encouragement and facilities in field and laboratory work extended to me by the late Sir J. C. Bose, F.R.S., the then Director and Dr. D. M. Bose, the present Director, Bose Research Institute, Calcutta, where the work has been carried out. My best thanks are also due to Mr. D. Mukerji, Lecturer in Zoology, Calcutta University, for valuable suggestions and as well as for the identification of the ants; to Dr. Gravely, Director, Madras Museum, for the identification of several specimens of spiders and to Dr. J. P. Sirkar and Mr. S. Sen-Gupta for generous help and suggestions.

References.

[1] Blackwall, J.—'Notice of several recent Discoveries in the Structure and Economy of Spiders', Trans. Linn. Soc., Lond., Vol. XVI, pt. 1, pp. 483-484, 1831.

[2] 'Researches into the Structure, Functions and Economy of the Araneides' (From the Report of the Meeting of the Association held at York, 1844), Ann. and Mag. of Nat. Hist., Vol. XV., p. 230., Lond., 1845.

[3] 'A History of the Spiders of Great Britain and Ireland', pt. 1, Intro. p. 7, Lond., 1860.

[4] Pollock, F.—'On the History and Habits of the Epeira aurelia Spider', Ann. and Mag. of Nat. Hist., Vol. XV, Ser. III, pp. 461-462, 1865.

[5] Wilder, B. G.—'The Habits and Parasites of Epeira riparia, with a note on the moulting of Nephila plumipes', Proc. Amer. Assoc. for Advancement of Science, p. 262, Aug. 1874.

[6] Wagner, M. Wol.—'La mue des Araignées', Ann. Sci. Nat. (Zool.) T. VI, pp. 281–389, Paris, 1888.

[7] McCook, H. C.—'American spiders and their spinning work', Vol. III, pp. 95–99, 1893.

[8] Simon, E.—'Histoire Naturelle Des Araignées' Déuxieme edition, Tome-premier, pp. 43–47, 1892–95.

[9] Dr. Leach.—'Annulosa'—Supplement to the Encyclopædia Britannica, p. 435.

[10] Pocock, R. I.—'Arachnida'—Fauna British India, p. 154, Lond, 1900.

[11] Comstock, J. H.—The Spider Book, pp. 183-184, N.Y., 1920.

[12] Warren, E.—'Note on the ecdysis of a Spider', Ann. Natal. Museum, Vol. V, pt. II, pp. 231–234, 1925.

[13] Fischel, W.—'Wachstum und Hautung der Spinnen', Zeits. für Wiss. Zool., Vol. 136, pp. 79–107, 1930.

[14] Bonnet, P.—'La mue l'autotomie et la régénération chez les Araignées', Bull. Soc. D'Hist. Nat. Toulouse. Tome. Cinquante neuvieme., (La mue, pp. 237–504). 65e Année., 1930.

[15] Millot, J.—'La métamérisation postembryonnaire des Jeunes Araignées,' Bull. De la Soc. Zool. de France. Cinquante-Sixieme Vol., Année., 1931.

[16] Savory, T. H.—The spiders and allied orders of the Brit. Isles, p. 10, 1935.

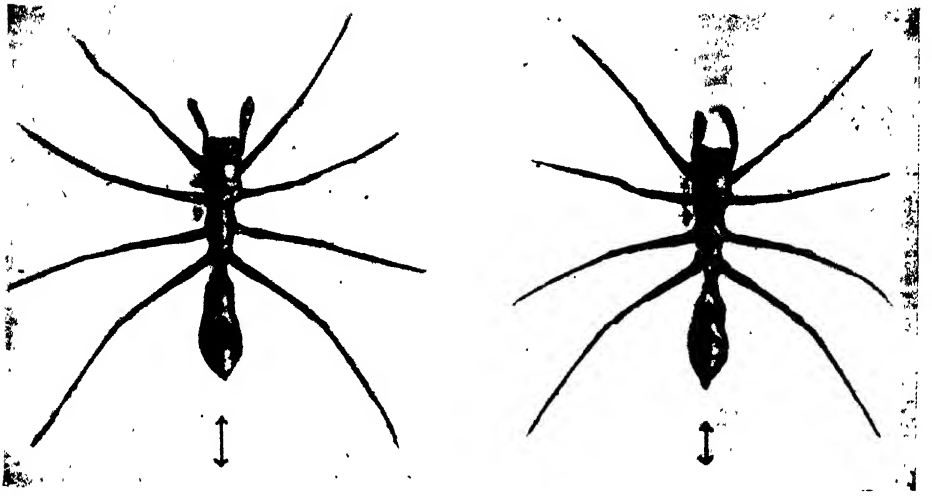
[17] Cambridge, O. P.—'Descriptions and sketches of some new species of Araneidea, with characters of new Genus' (Salticus plataleoides, N. Sp.), Ann. and Mag. Nat. Hist., Vol. III, Ser. IV, pp. 68-69, 1869.

[18] Peckham, G. W. and E. G.—'Ant-like spiders of the family Attidae', Occasional papers of the Nat. Hist. Soc. of Wisconsin, Vol. II, pp. 33-34, 1892.

[19] Pollock, F.—'On the History and Habits of E. aurelia spider', Ann. and Mag. Nat. Hist., Vol. XV, Ser. III, p. 461, 1865.

[20] Simon, E.—Hist. Nat. des Araignées, Deuxieme edition. Tome—Second, pp. 502-3, 1897–1901.

[21] Peckham, G. W. and E. G.—'Ant-like spiders of the family Attidae', Occasional papers of the Nat. Hist. Soc. of Wisconsin, Vol. II, pp. 33-34, 1892.
[22] Hingston, R. W. G.—'Field observations on spider mimics', Proc. Zool. Soc., Lond., pt. II, p. 844, 1927.
[23] Narayan, K.—'Notes on Ant-like spiders of the family Attidae', Rec. Ind. Mus., Vol. XI, p. 396, 1915.
[24] Mukerjee, D. D.—'Report on a collection of ants', Jour. Bom. Nat. Hist. Soc., Vol. XXXIV, (i), pp. 157-158, 1930.
[25] Mathew, A. P.—'The Life History of the spider M. plataleoides', Jour. Bom. Nat. Hist. Soc., Vol. XXXVII, pp. 371–73, 1934.
[26] Bhattacharya, G.—'The fish-eating Spiders in Bengal and their habits', Trans. Bose Inst., Vol. VII, p. 246, 1931-32.
[27] Pollock, F.—'On the History and Habits of E. aurelia spiders', Ann. and Mag. Nat. Hist., Vol. XV, Ser. III, p. 461, Lond., 1865.
[28] Emerton, J. H.—'New England spiders of the family Attidae', Trans. Connecticut Academy of Arts and Science, New Haven, Vol. VIII, p. 221, 1888–1892.
[29] Bristowe, W. S.—'The mating habits of British Thomisid and Sparassid spiders', Ann. and Mag. Nat. Hist., Vol. XVIII, Ser. IX, pp. 127-128, 1926.
[30] Blackwall, J.—'The Structure and Economy of Spiders', Trans. Linn. Soc., Lond., Vol. XVI, pt. 1, p. 484, 1831.
[31] Comstock, J. H.—The Spider Book, p. 184, N.Y. 1920.
[32] Wagner, M. Wol.—'La mue des Araignées', Ann. Sci. Nat. (Zool.) Tom VI, p. 371, 1888.
[33] McCook, H. C.—American Spiders and their spinning work, Vol. I, p. 24, 1889.
[34] Darwin, C.—The Descent of man and selection in relation to sex. Edtn. 2nd., pt. II, 272, 1874.
[35] Simon, E.—Hist. Nat. des Araignées, Premier Partie., p. 46, 1892–95.
[36] Warburton, C.—'Arachnida Embolobranchiata', Cambridge Nat. Hist., Vol. IV, p. 338, 1909.
[37] Staveley, E. F.—British Spiders, p. 22, Lond., 1866.
[38] Blackwall, J.—Spiders of Great Britain and Ireland, pt. I, Intro. p. 7, Lond., 1860.
[39] McCook, H. C.—American Spiders and their spinning work, Vol. II, p. 206, 1890.
[40] Wagner, M. Wol.—'La mue des Araignées', Ann. Sci. Nat. (Zool.) T. VI, p. 371, Ser. VII, Paris, 1888.
[41] Simon, E.—Histoire Naturelle des Araignées, Tome Premier, p. 43, 1892–1895.



(*a*) (*b*)

FIG. 1. Enlarged photographs of immature specimens of *Myrmarachne plataleoides*, Camb.

(*a*) The potential male of *M. plataleoides*. (Photograph taken after the 7th day of the 5th moult.)

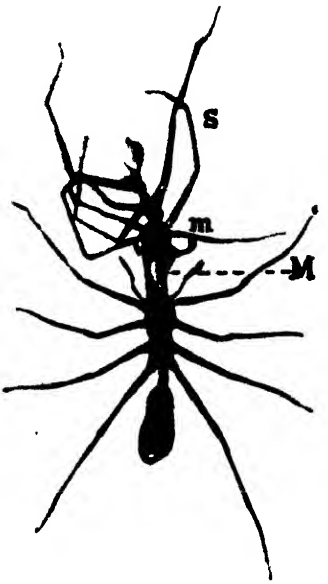
(*b*) The potential female of *M. plataleoides*. (Photograph taken after the 15th day of the 5th moult.)

(*a*)

FIG. 2. Enlarged photograph of different stages of the final moult of the potential male of *M. plataleoides*.

(*a*) The old cephalothoracic cuticle 'C' is lifted off like a lid. The growing chelicerae 'M' are exposed in order to have a clear view. Old mandibular cuticles 'm' are seen lying in front.

(*b*)



(*c*)

FIG. 2.

(*b*) Further progress of moulting of the potential male. The spider is drawing out its first and second pair of legs from the slough 'S'.

(*c*) The spider is disengaged from the exoskeleton in a wrong way. The slough 'S' dangles from the growing chelicerae 'M'. The old mandibular cuticle 'm' is seen attached to them.

(*d*)

(*d*) The spider has completely cast off its skin; 'S'—the cast off skin; 'C'–the cephalothoracic cuticle. Note the minute mandibular cuticle 'm' in the cast off skin.

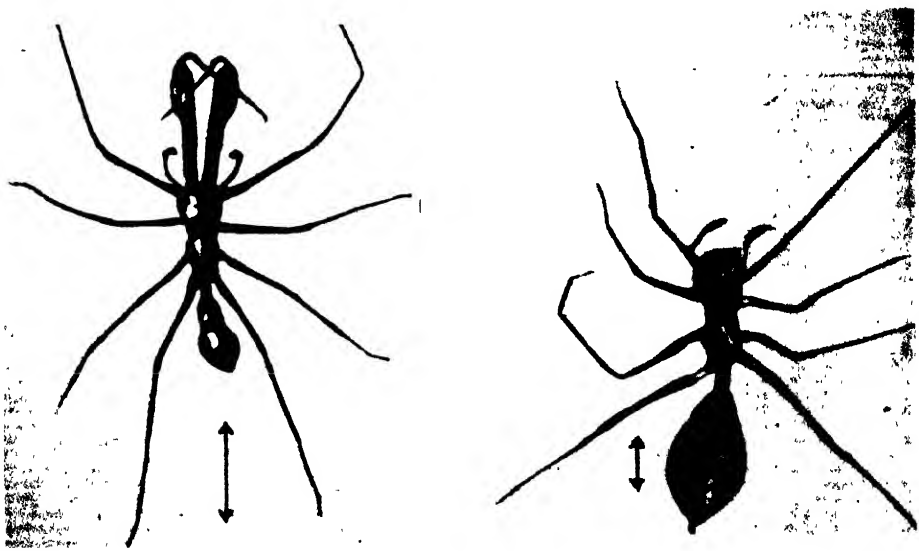
FIG. 3.

(*a*) External structure of the pedipalp of sex-undetermined specimen and adult female of *M. plataleoides.*

(*b*) The pedipalp of the male after the final moult.

(*c*) The chelicera of the sex-undetermined specimen and adult female of *M. plataleoides.*

(*d*) The chelicera of male which grows out of a minute one as shown in 'C' during the final moult.



(*a*) (*b*)

FIG. 4.

(*a*) Enlarged photograph of the adult *M. plataleoides*, Camb. ♂.

(*b*) Enlarged photograph of the adult *M. plataleoides*, Camb. ♀.

XI. ON THE DISPERSION OF SUPERSONIC WAVES IN A LIQUID.

By A. K. DUTTA, D.Sc.

(*Received 10th February, 1938.*)

It has been definitely established, for some time past now, that the velocity of sound waves in a gaseous medium does not remain constant throughout the range of frequencies. It has been experimentally observed that the general nature of the relationship between the frequency and the velocity can be depicted by a curve like that shown in fig. 1. Such a dispersion in gases has been explained on theoretical grounds.

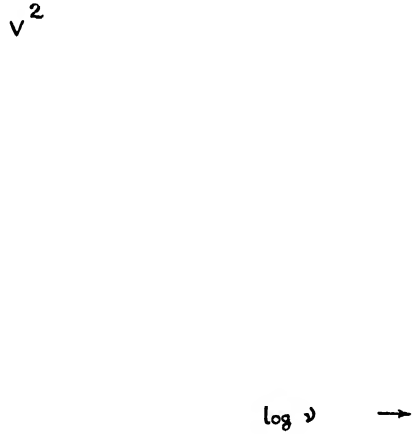


FIG. 1.

From the nature of the curve one can easily see that the velocity remains constant for a range of frequencies beginning with a very low frequency of sound waves. Then, for a particular region of frequency the velocity begins to increase with the frequency and comes again to a stationary value after a limited region. The particular range of frequency, at which this increase of velocity becomes apparent, is a characteristic of the substance. It has been observed that the dispersion of sound waves in a gaseous medium begins, generally, with high

frequencies of the order of 10^5 cycles per second. In the case of liquids, such a dispersion of sound waves has not yet been definitely established, nor has any dispersion formula of sound waves with frequency been worked out *. The problem of the present paper is to work out fully a dispersion formula in the case of liquids and to trace out the expected behaviour in the case of different liquids. The general method of procedure has been as in the case of gases, and to generalise where the specific gas laws have been applied.

Herzfeld and Rice [1], and Kneser [2] gave an explanation for the dispersion of sound waves in gases. The fundamental ground for the change of velocity with frequency was considered to be a time lag between the energy impressed by the sound waves and the internal energy of the medium as embodied in the oscillatory motions. The general thermodynamic relationship,

$$CdT+pdv=0 \quad .. \quad .. \quad .. \quad (1)$$

can be written as

$$C_a dT+C_i' dT+pdv=0 \quad .. \quad .. \quad (2)$$

where $C_i' dT$ denotes the change in energy due to the assimilation in the internal degrees of freedom and $C_a dT$ denotes the change in energy in other possible ways, like translatory motion, etc. The term $C_i' dT$ can be considered as a function of frequency and should be written as $C_i' dT = C_i F(\omega) dT$, with the condition that $F(0) = 1 > F(\omega) > 0 = F(\infty)$.

The equation (2) can, then, be written more explicitly as,

$$C_a dT+C_i F(\omega) dT+pdv=0 \quad .. \quad .. \quad (3)$$

The velocity of sound is conveniently expressed by the relationship,—

$$V_{\text{com}}^2 = \frac{dp}{d\varrho} = \frac{\partial p}{\partial v} \cdot \frac{\partial v}{\partial \varrho} = \frac{p}{\varrho}\left(1 - \frac{1}{p}\frac{d(pv)}{dv}\right) \quad .. \quad (4)$$

$$\left\{\text{since, } \frac{\partial p}{\partial v} \cdot \frac{\partial v}{\partial \varrho} = \left(\frac{d(pv)}{dv} - p\right)\frac{1}{v} \cdot \frac{\partial v}{\partial \varrho} = \frac{1}{v} \cdot \left[\frac{(dpv)}{dv} - p\right] \cdot \frac{-v}{\varrho}\right\}.$$

* Kneser (Ann. der Phys., 32, 277, 38) has since worked out a general dispersion formula in the case of liquids. The derivation in this paper comes out as a particular case with known constants.

If, in the above equation, we substitute the value of pdv from equation (3) and put $d(pv) = RdT$, we obtain,

$$V^2_{com} = \frac{p}{\rho}\left(1+\frac{RdT}{C_a dT + C_i F(\omega) dT}\right)$$

$$= \frac{p}{\rho}\cdot\left(1+\frac{R}{C_a + C_i F(\omega)}\right) \quad .. \quad .. \quad (5)$$

In the case of liquids, however, we can use the relation,

$$H = E + pv$$

so that $$dH - dE = d(pv)$$

or $$C_p dT - C_v dT = d(pv).$$

Hence, $$d(pv) = \frac{\alpha^2 V_m T}{\chi}\, dT \quad .. \quad .. \quad .. \quad (6)$$

where α and χ are the coefficients of expansion and compressibility and V_m is the molar volume. Hence, in the case of liquids we should have the relation,

$$V^2_{com} = \frac{p}{\rho}\left(1+\frac{\alpha^2 V_m T/\chi}{C_a + C_i F(\omega)}\right) \quad .. \quad .. \quad (7)$$

[The function $F(\omega)$ can be derived to be equal to $\frac{1}{1+j\omega\beta}$ as follows :—

The equation

$$C_a dT + C_i F(\omega) dT + pdv = 0$$

can be written in the form

$$C_a dT + E_1 \Delta n_1 + pdv = 0$$

where E_1 is the energy taken up by each molecule and Δn_1 is the change in the number of excited molecules. Hence,

$$C_i F(\omega) = \frac{E_1 \Delta n_1}{dT}$$

But $$-\frac{\partial n_1}{\partial t} = k_1 n_1 - k_0 n_0$$

where k_1 and k_0 give the number of transitions per unit of time from oscillatory energy to general energy and vice versa. For harmonic disturbance, we have

$$-j\omega n_1 = k_1 n_1 - k_0 n_0$$

or $$-j\omega \Delta n_1 = k_1 \Delta n_1 - k_0 \Delta n_0 + n_1 \Delta k_1 - n_0 \Delta k_0$$

But $\Delta n_0 = -\Delta n_1$ (since we are considering only one particular excited state).

Hence, $$-j\omega \Delta n_1 = k_1 \Delta n_1 + k_0 \Delta n_1 + n_1 \Delta k_1 - n_0 \Delta k_0$$

or $$\Delta n_1 \cdot \frac{n_0 \Delta k_0 - n_1 \Delta k_1}{j\omega + k_1 + k_0}$$

and $$\Delta n_1 (\omega = 0) = \frac{n_0 \Delta k_0 - n_1 \Delta k_1}{k_1 + k_0}$$

we have then,

$$\frac{C_i F(\omega)}{C_i F(0)} = \frac{k_1 + k_0}{j\omega + k_1 + k_0} = \frac{1}{1 + \frac{j\omega}{k_1 + k_0}} = \frac{1}{1 + j\omega\beta}$$

where $\beta = \frac{1}{k_1 + k_0}$.

Since $F(0) = 1$, we have

$$F(\omega) = \frac{1}{1 + j\omega\beta} \cdot \Big]$$

Considering the real and imaginary parts of equation (7) and substituting the value of $F(\omega)$ we have the real part given by

$$V^2 = \frac{p}{\varrho}\left(1 + \frac{\alpha^2 V_m T}{\chi} \cdot \frac{C + \omega^2 \beta^2 C_a}{C^2 + \omega^2 \beta^2 C_a^2}\right) \quad .. \quad (8)$$

where $$C = C_a + C_i \quad .. \quad .. \quad .. \quad (9)$$

As in the case of gases[3], we have then,

$$\frac{dV^2}{d \log \omega} = \frac{p}{\varrho} \frac{2\alpha^2 V_m T}{\chi} \cdot \frac{C \cdot C_i C_a \omega^2 \beta^2}{(C^2 + \omega^2 \beta^2 C_a^2)^2} \quad .. \quad (10)$$

which, when plotted, gives a curve of the type of fig. 1.

We have further

$$V_0^2 = \frac{p}{\varrho}\left(1+\frac{\alpha^2 V_m T}{\chi}\cdot\frac{1}{C}\right) \quad .. \quad .. \quad (11)$$

$$V_\infty^2 = \frac{p}{\varrho}\left(1+\frac{\alpha^2 V_m T}{\chi}\cdot\frac{1}{C_a}\right) \quad .. \quad .. \quad (11a)$$

so that $$\frac{V_\infty^2}{V_0^2} = \frac{C_a+\alpha^2 V_m T/\chi}{C_a}\times\frac{C}{C+\alpha^2 V_m T/\chi} \quad .. \quad (11b)$$

It is thus evident, that whenever we can find out the magnitudes of C and C_a individually and the coefficients α and χ, we can have a measure of the ratio of the two velocities V_0 and V_∞ even in the case of liquids and if we can have an estimate of the value of $\beta = \frac{1}{k+k_0}$ it is not even difficult to draw out the full curve of dispersion in the cases of various liquids.

In order to calculate the values of C, C_a, etc. one has to make use of the data given by Raman effect and the infrared spectra for the frequency of oscillation. We can calculate the value of C_i given by the Einstein function as

$$C_i = \frac{\theta^2 e^\theta}{(e^\theta-1)^2} \text{ (R units) where } \theta = h\nu/kT \quad .. \quad (12)$$

Hence, calculating the value of C_i we can find out the values of C_a from the known values of C, the specific heat at constant volume. Thus in the cases where we can pick up the proper forms of oscillation energies involved, we can calculate the ratio of $V_\infty : V_0$, provided other data are also known. The following table with different liquids has been worked out on this hypothesis. The oscillation frequencies considered in the table are taken from the Raman effect values and generally, only the ground oscillations have been taken. The values of the frequencies and that of the other constants involved have been taken from Landolt and Börnstein tables and from the international critical tables.

TABLE

Liquid.	Frequency Cm^{-1}	$sec^{-1} \times 10^{13}$	$\theta = h\nu/kT$ at 25°C	$C_i = \frac{\theta^2 e^\theta}{(e^\theta - 1)^2}$ (R units)	$\frac{\alpha^2 V_m T}{\chi} = C_p - C_v = k'$ (calories)	C_p in calories	C_v in calories	$\frac{V_\infty^2}{V_0^2} = \frac{C_v - C_i + k'}{C_v - C_i} \times \frac{C_v}{C_v + k'}$
Toluol ..	217	0·65	1·04	·915	11·6	38	26·4	1·022
	730	2·2	3·5	·39				1·01
	2730	8·2	13·1	·00036				1·000....
Xylol ..	733	2·2	3·5	·39	11·2	42	30·8	1·008
	2730	8·2	13·1	·00036				1·000....
Aniline ..	756	2·3	3·68	·36	10·6	47	36·4	1·004
	3360	10·0	16·0	·00003				1·000....
Water ..	1652	4·9	7·8	·027	·045	18	17·95	1·000....
	3420	10·2	16·3	·00002				1·000....
	3580	10·7	17·1	....				1·000....
Carbon-bi-sulphide	395	1·00	1·6	·812	6·5	18·1	11·6	1·057
	656	1·97	3·1	·476				1·032
	796	2·4	3·8	·338				1·022
Carbon-tetra-chloride	218	0·64	1·04	·915	9·4	31·2	21·8	1·027
	314	0·94	1·50	·83				1·024
	459	1·37	2·2	·68				1·020
	759	2·28	3·64	·37				1·011

From a perusal of the table it becomes apparent that except in the case of water, a dispersion effect of the total magnitude varying from one to five per cent of the V^2 value is expected in all other liquids considered. This magnitude is evidently a measurable quantity in the present condition of the technique, provided it lies in the supersonic frequency region. In many of the liquids considered there are different dispersion effects according to the frequency of oscillations considered and as in the cases of gases it is also likely that these different dispersion effects fall in different regions of frequencies. It is, however, not beyond the range of possibility that two or more dispersion effects overlap in the same region of frequency and thus give a complicated effect. Taking, however, any particular case of a liquid and a particular frequency of oscillation we can trace out the full nature of the dispersion curve, although the region of frequencies at which the curve lies is yet difficult to foretell. As an example, we can take the case of Toluol with the value of $V_0 = 1{,}304$ meters per sec. at 25°C and the frequency of oscillation that gives rise to dispersion to be 730 cm^{-1}. Combining the formulæ (8) and (11) we can write

$$V^2 = V_0^2 \frac{1}{1+\frac{\alpha^2 V_m T}{\chi . C}} \left\{ 1+\frac{\alpha^2 V_m T}{\chi} \cdot \frac{C+\omega^2\beta^2 C_a}{C^2+\omega^2\beta^2 C_a^2} \right\} \quad .. \quad (13)$$

Excepting the values of $\omega\beta$ all the other values in the above equation are known. Giving $\omega\beta$ all possible values from 0 to ∞, we obtain the values of V^2 for different values of $\omega\beta$ as shown below:

$\omega\beta$	$= 0$	·1	1	10	∞
$V^2 \times 10^6 =$	1·701	1·701	1·708	1·718	1·718

The values are correct to the order of one per thousand.

We can trace out the curve of V^2 against log $\omega\beta$ from the values given above, and as will be evident from the figure, the curve is exactly of the same type as obtained for the gases.

Since β is a constant quantity for a particular substance, the curve depicts also the relationship between V^2 and log ω.

But the exact values of ω are however unknown. From the curve we can read that the region of dispersion lies in the ω

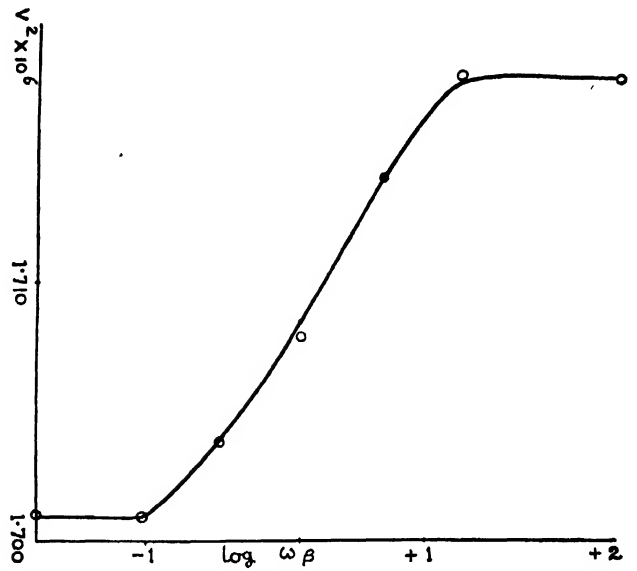


FIG. 2.

values given by $\log \omega_0 - \log \omega_\infty = -2$. It can however be surmised, since the packing of the molecules in the liquid state is about 1,000 times the packing in the gaseous state, the corresponding frequency for dispersion will be 10^3 times higher in the case of liquids than in the case of gases. The region of frequency thus comes to be of the order of 10^8 or 10^9, which is yet beyond the range of experimental supersonics.

As regards the experimental investigations on the supersonic dispersion in liquids, Spakovskij[4] and Parthasarathy[5] had obtained negative results with the liquids tried by them and Hiedemann[6] with his co-workers had reported effect of 1 in 1000 in the cases of Toluol and Xylol and no dispersion in the case of water. (After a colloquium in Berlin in the year 1937, we had a discussion with Hiedemann on the point. He expressed his personal conviction that he could not rely on the values published by him due to various reasons.) So far, then, up to the end of 1937, no reliable experimental data on supersonic dispersion was available, and the expected behaviour of different liquids on the basis of the gas dispersion theory had not been

worked out. After completing this work on the expected behaviour of the liquids, I had undertaken to investigate experimentally the dispersion of supersonic waves in different liquids, the results of which have since been published[7].

My thanks are due to Prof. Debye for suggesting the problem and to the authorities of the Bose Institute for granting me study leave for an year which made it possible for me to carry out this work in Germany.

In conclusion I beg to thank Prof. D. M. Bose for his valuable discussions before sending it to the press.

REFERENCES.

[1] Herzfeld and Rice.—Phys. Rev., 31; 691; 1928.
[2] Kneser.—Ann. der Physik, 11; 761, 777; 1931.
[3] Hiedemann.—Ergebnisse der Exakt. Naturwiss, 14, 201, 1935.
[4] Spakovskij.—C.R., Leningrad, 3, 588, 1934.
[5] Parthasarathy.—Proc. Ind. Acad. Sci., 4, 17, 1935.
[6] Hiedemann, Seifen, and Scheurer.—Naturwiss, 24, 681, 1936.
[7] Dutta.—Phys. Zeits., 39, 186, 1938.

XII. ON THE REFLECTION OF ELECTROMAGNETIC WAVES IN THE IONOSPHERE.*

By R. C. MAJUMDAR.

(*Received 5th November 1938.*)

ABSTRACT.

The conditions for the total reflection of electromagnetic waves in the ionosphere are examined directly from the definition that the total reflection takes place when the flow of energy associated with the wave vanishes across the wave front. It is found that the electromagnetic waves are totally reflected from a layer where either the refractive index vanishes or it becomes infinitely large. The critical penetration frequencies are worked out in general when the damping of the wave is not neglected. The Sellmeyer vs. Lorentz theory is also discussed in this connection.

INTRODUCTION.

It has been shown in a series of recent papers [1] that if a proper programme of theoretical research is carried out to investigate the mechanism of absorption of the electromagnetic waves in upper atmosphere and therefrom to evaluate the expression for collision frequency of the electrons with gas particles,† supposed to be present in the Ionosphere, then by comparing these theoretical results with those obtained from the measurement of the reflection coefficient or from the process of interaction of radio waves, one can obtain valuable informations as regards the constituents and the fundamental mechanism in the ionosphere. For the purpose we developed a general quantum Kinetic theory for investigating the phenomena of Dispersion, Absorption and polarisation of radio waves in Ionosphere, which is to be substituted for that of Appleton-Hartree's [2] usual procedure if

* The paper was read in the meeting of the Indian Physical Society on the 30th April, 1938.

† The particles may be neutral or ionised atom and molecules.

we want to find out a formula for the absorption coefficient or the collision frequency of the electrons, which can be really compared with the observations. But the question under which conditions the electromagnetic waves get totally reflected in the upper atmosphere, a fact which can also be investigated experimentally and offers valuable informations as to the structure of the Ionosphere, was left open in those previous works. The usual procedure as adopted by Appleton[3] and others has been to put the refractive index of the medium equal to zero as the condition of the total reflection of the radio waves; the three reflections, one for ordinary and two for extraordinary waves, which have been observed by various workers when the frequency of the radio wave is much greater than the gyrofrequency are in agreement with this postulate of Appleton. Recently, however, the radio workers at Allahabad[4] have discovered one more reflection in addition to the three just mentioned above and this is attributed to the extraordinary wave and obtained *a priori* as first shown by Goubau[5] and afterwards by Rai[6], if we assume that the total reflection takes place when the group velocity of the wave train vanishes. These investigations, therefore, call for a closer theoretical analysis of the criterion of total reflection of radio waves in the upper atmosphere. The present investigation, which is devoted to this analysis from the fundamental principle of energy propagation associated with the electromagnetic waves, is the outcome of a stimulating discussion of the author with Professor S. N. Bose to whom he is greatly indebted for having the opportunity of reading his recent paper[7] in Manuscript. He is also grateful to Dr. D. M. Bose, the Director of the Institute, for his interest and encouragement.

§1. *Conditions for total reflection.*

We start with the recapitulation of some of the properties of Maxwell's equations as well as of the procedure we adopted in previous papers.[1]

$$\left.\begin{aligned} &\text{rot } \vec{H} = \frac{1}{c}\frac{dD}{dt}, \quad \text{rot } \vec{E} = -\frac{1}{c}\frac{\partial H}{\partial t} \\ &\text{div } \vec{E} = 4\pi\varrho, \quad \text{div } \vec{H} = 0. \end{aligned}\right\} \tag{1}$$

where the vectors $\vec{H}$, $\vec{D}$, $\vec{E}$, etc. have usual meaning and are proportional to

$$e^{iw\left(t-\frac{n}{c}\left(\vec{r}\,\vec{s}\right)\right)},$$

$\vec{s}$ being the unit vector in the direction of wave normal and n the refractive index. The first two of the Maxwell's equations are thus reduced to

$$n\left[\vec{H}\vec{s}\right]=\vec{D} \text{ and } n\left[\vec{E}\vec{s}\right]=-\vec{H} \quad .. \quad .. \quad (2)$$

or eliminating $\vec{H}$ from these equations we obtain

$$\vec{D}=n^2\left\{\vec{E}-\vec{s}\left(\vec{E}\vec{s}\right)\right\} \quad .. \quad .. \quad (3)$$

We therefore find that H stands at right angles to the vectors $\vec{D}$, $\vec{E}$, and $\vec{s}$, which are coplanar, $\vec{D}$ being perpendicular to the vector $\vec{s}$, which denotes the direction of propagation. It is

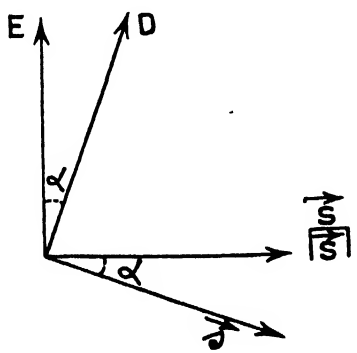


FIG. 1.

to be noted that the direction of propagation is not in the same direction as that of the energy transport but makes an angle α with it.

Now the displacement vector $\vec{D}$ is defined by

$$\vec{D}=\varepsilon^{i}.\vec{E} \quad .. \quad .. \quad .. \quad (4)$$

where the dielectric tensor ε^t is determined by solving in the usual way the Boltzmann's integral equation in electric and magnetic field. It is thus obtained

$$\varepsilon^t = U^t + K'.\ U^t - L'.\left[U^t\vec{\omega}_L\right] + M'.\vec{\omega}_L\vec{\omega}_L \quad .. \quad (5)$$

where U^t is the unit tensor and K', L' and M' are the integral tensors, evidently of diagonal character and are given in the previous papers.

We thus obtain finally the determinantal tensor equation *

$$\left(\varepsilon^t - n^2U^t + \vec{n}\vec{n}\right).\vec{E} = 0, \quad .. \quad .. \quad (6)$$

the elimination of E_x, E_y, E_z from which gives us an equation for the evaluation of the refractive index. The equation (6) itself then gives the ratios $E_x : E_y : E_z$, i.e. the nature of polarisation for the corresponding wave in the medium.

We have further the following important relations [8] which can be easily verified.

The total energy density is given by

$$W = \frac{1}{8\pi}\left\{\left(\vec{E}\vec{D}\right) + \vec{H}^2\right\} = \frac{n^2}{4\pi}\left\{\vec{E}^2 - \left(\vec{E}\vec{s}\right)^2\right\}, \quad .. \quad (7)$$

whereas the flow of energy as determined by the Poynting's vector is

$$\vec{S} = \frac{c}{4\pi}\left[\vec{E}\vec{H}\right] = \frac{cn}{4\pi}\left\{\vec{s}\vec{E}^2 - \vec{E}\left(\vec{E}\vec{s}\right)\right\} \quad .. \quad (8)$$

and further

$$\left(\vec{S}\vec{s}\right) = \frac{cn}{4\pi}\left\{\vec{E}^2 - \left(\vec{E}\vec{s}\right)^2\right\} = vW \quad .. \quad (9)$$

$$v = \text{velocity of the wave front} = \frac{c}{n} \quad .. \quad (10)$$

Let us now discuss the condition of total reflection. The proper way of approach will be to consider the whole problem

* This is equivalent to the fundamental equation (B) in Prof. S. N. Bose's paper. It can be easily verified by writing S for the phase of the wave, i.e. $\vec{E}$, $\vec{D}$, $\vec{H}$ are proportional to e^S. This method will be particularly useful when we want to investigate the properties of the wave that will leak through the barrier of total reflection.

in terms of propagation of energy associated with the electromagnetic waves. It is a well-known fact in the electromagnetic theory that in the case of total reflection the energy flows along the boundary surface of separation but does not enter the second medium (we neglect thereby the leakage through the boundary wall). Thus for total reflection we must have $(\vec{S}\vec{s}) = 0$ and this is satisfied as obvious from (9) and (7) either

(i) the refractive index $n \to 0$

or (ii) the refractive index $n \to \infty$.

It can also be easily verified that for these two cases the energy of the incident wave is simply equal to the energy of reflection, the refracted energy being zero. We have, as for example, for normal incidence the Reflection Coefficient, i.e. the ratio of the reflected to that of incident energy

$$R = \frac{|n-1|^2}{|n+1|^2} \quad .. \quad .. \quad .. \quad (11)$$

and the Transmission Coefficient (Durchlässigkeit Koeffizienten), i.e. the ratio of the refracted to that of the incident energy

$$D = \frac{4n}{|n+1|^2} \quad .. \quad .. \quad .. \quad (12)$$

Thus for $n \to 0$ or $n \to \infty$ we obtain $R = 1$, $D = 0$, showing that the wave is totally reflected.

The following diagram[9] shows difference between the two conditions of total reflection :

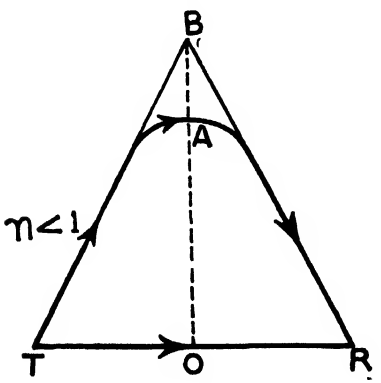


FIG. 2a.

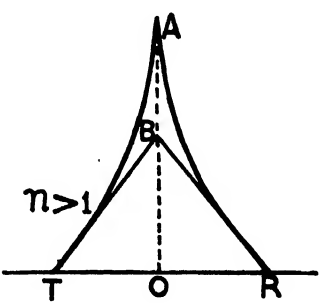


FIG. 2b.

We therefore find that for the first case the equivalent height OB is greater than the actual height OA of the Ionosphere whereas reverse is the case for the second one.

§ 2. *Penetration frequency and the Electron density.*

For the propagation of plane electromagnetic waves in the z-direction, the external earth's field being resolved into two components, H_L along the direction of propagation and H_T at right anlges to it, we obtain the refractive index for the normal incidence $\vec{n}=(0, 0, \mu)$

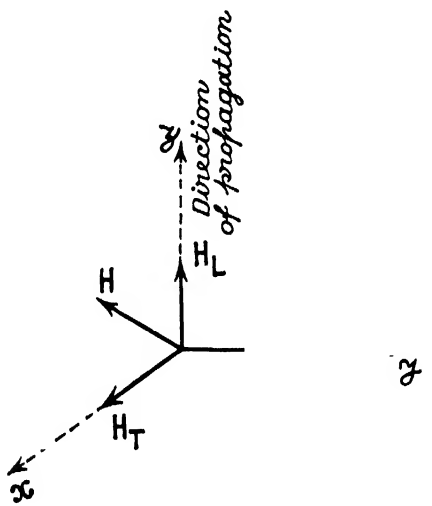


FIG. 3.

$1-\mu^2_{\text{complex}}$

$$\left(1-\frac{i\bar{\Delta}^n}{\omega}\right)-\frac{y_T^{\,2}}{2\left(1-x_0-\frac{i\bar{\Delta}^n}{\omega}\right)}\mp\frac{1}{2\left(1-x_0-\frac{i\bar{\Delta}^n}{\omega}\right)}\sqrt{y_T^{\,4}+4y_L^{\,2}\left(1-x_0-\frac{i\bar{\Delta}^n}{\omega}\right)^2}$$

x_0

and

$$\frac{H_y^0}{H_x^0}=\frac{iy_L(1-\mu^2)}{\left(1-\frac{i\bar{\Delta}^n}{\omega}\right)(1-\mu^2)-x_0} \quad .. \quad .. \quad (14)$$

where

$$\bar{\Delta}^n = \int_0^\infty \Delta^n(\varepsilon)\varepsilon^{\frac{1}{2}}\frac{\partial f_0}{\partial \varepsilon}\,d\varepsilon \Big/ \int_0^\infty \varepsilon^{\frac{1}{2}}\frac{\partial f_0}{\partial \varepsilon}\,d\varepsilon \quad .. \quad (15)$$

In the particular case when the ionosphere consists of electrons and ions only the expression (15) is reduced to

$$\bar{\Delta} = \frac{4\pi}{3}\frac{e^4N^+}{(2\pi mk^3)^{\frac{1}{2}}}\frac{1}{T^{\frac{3}{2}}}\left\{\log(kT\alpha) - 1{,}577\right\} \quad .. \quad (16)$$

$$= 1{,}8\,\frac{N^+}{T^{\frac{3}{2}}}\left\{\log(kT\alpha) - 1{,}577\right\} \quad .. \quad .. \quad (17)$$

Here

$$x_0 = \frac{4\pi Ne^2}{m\omega^2}, \qquad y_{L,T} = \frac{eH_{L,T}}{mc\omega}, \qquad \omega_H = \frac{eH}{mc},$$

$$\frac{\omega}{2\pi} = \nu \text{ (frequency of the wave)}$$

and

$$\alpha = 6{,}63\,10^{27}b^2 \quad .. \quad .. \quad .. \quad .. \quad .. \quad (18)$$

where b = screening constant, N = number of electrons in unit volume, N^+ = number of ions in unit volume, and H = earth's magnetic field.

From the condition postulated above we obtain evidently the following equation to be satisfied when the total reflection takes place and which gives the desired relation between the penetration frequency and the electron density. We have, writing $\bar{\Delta}$ for $\bar{\Delta}^n$ for convenience,

$$\left(1-\frac{i\bar{\Delta}}{\omega}\right) - \frac{y_T^2}{2\left(1-x_0-\frac{i\bar{\Delta}}{\omega}\right)}$$

$$\mp \frac{1}{2\left(1-x_0-\frac{i\bar{\Delta}}{\omega}\right)}\sqrt{y_T^4 + 4y_L^2\left(1-x_0-\frac{i\bar{\Delta}}{\omega}\right)} = x_0 \quad .. \quad (19)$$

$$= 0 \quad .. \quad (20)$$

according as $\mu = 0$ or $\mu = \infty$.

Solving (19) we obtain

either $$1-\frac{i\bar{\Delta}}{\omega}-x_0=0 \quad .. \quad .. \quad .. \quad (21)$$

or $$1-\frac{i\bar{\Delta}}{\omega}-x_0=\pm y \quad .. \quad .. \quad (22)$$

i.e. either $$\omega^2-i\bar{\Delta}\omega-\frac{4\pi Ne^2}{m}=0 \quad .. \quad .. \quad .. \quad (23)$$

or $$\omega^2-\omega(i\bar{\Delta}\pm\omega_H)-\frac{4\pi Ne^2}{m}=0 \quad .. \quad .. \quad (24)$$

From which follows immediately

either $$\omega=\sqrt{\frac{4\pi Ne^2}{m}-\frac{\bar{\Delta}^2}{4}}+\frac{i\bar{\Delta}}{2} \quad (25)$$

or $$\omega=\sqrt{\frac{4\pi Ne^2}{m}+\frac{\omega_H^2}{4}-\frac{\bar{\Delta}^2}{4}}\pm\frac{\omega_H}{2}$$

$$+\frac{i\bar{\Delta}}{2}\left\{1\pm\frac{\omega_H}{2}\Big/\sqrt{\frac{4\pi Ne^2}{m}+\frac{\omega_H^2}{4}-\frac{\bar{\Delta}^2}{4}}\right\} \quad .. \quad (26)$$

In choosing the sign before the radical we assume that the characteristic frequency of the medium $\left(\frac{4\pi Ne^2}{m}\right)^{\frac{1}{2}}$ is greater than the frequency of the collision $\frac{\bar{\Delta}}{2}$ and retain therefore the positive sign only in order that the wave maintains its oscillatory character. As we are interested only in the real part of the frequency, the imaginary part gives simply the damping of the wave, we obtain the critical penetration frequencies at which the total reflections take place

$$\omega_C=2\pi\nu_C=\sqrt{\frac{4\pi Ne^2}{m}-\frac{\bar{\Delta}^2}{4}} \quad .. \quad (27)$$

$$\omega_{C\pm}=2\pi\nu_{C\pm}=\sqrt{\frac{4\pi Ne^2}{m}+\frac{\omega_H^2}{4}-\frac{\bar{\Delta}^2}{4}}\pm\frac{\omega_H}{2} \quad .. \quad (28)$$

whereas from (20) we obtain

either $$1 - \frac{i\bar{\Delta}}{\omega} - x_0 = 0 \quad .. \quad .. \quad .. \quad (21)$$

a relation which we have just studied,

or $$\left(1 - \frac{i\bar{\Delta}}{\omega}\right)^3 - x_0\left(1 - \frac{i\bar{\Delta}}{\omega}\right)^2 + x_0 y_L^2 - \left(1 - \frac{i\bar{\Delta}}{\omega}\right) y^2 = 0 \quad .. \quad (29)$$

From which follows immediately the relation between the penetration frequency and the electron density

$$\omega = \frac{4\pi e^2}{m} N. \frac{(\omega - i\bar{\Delta})^2 - \omega_L^2}{(\omega - i\bar{\Delta})^3 - (\omega - i\bar{\Delta})\omega_H^2} \quad .. \quad (30)$$

We do not however enter into the discussion of the solution for ω which will be complicated. We are much more interested in the relation between the penetrating frequency of the wave and the electron density which we obtain when we neglect the damping. Thus from (23) and (24) and (30) follows:

$$N = \frac{\pi m}{e^2} \nu_C^2 \quad .. \quad .. \quad .. \quad .. \quad (31)$$

$$N = \frac{\pi m}{e^2} \nu_C^2 \left(1 \pm \frac{\nu_H}{\nu_C}\right) .. \quad .. \quad .. \quad (32)$$

$$N = \frac{\pi m}{e^2} \nu_C^2 \frac{\nu_H^2 - \nu_C^2}{\nu_L^2 - \nu_C^2} \quad .. \quad .. \quad .. \quad (33)$$

where $$\nu_H = \frac{eH}{2\pi mc}, \; \nu_L = \frac{eH_L}{2\pi mc} .. \quad .. \quad .. \quad (34)$$

§3. *Sellmeyer vs. Lorentz theory.*

For completeness we add in this section a discussion on the relations between the penetration frequencies and electron concentrations when the force acting on the electron by electric field of the electromagnetic wave is not only the Maxwellian electric intensity E (the Sellmeyer theory) but also a force $\frac{4\pi}{3}P$ added due to the polarisation of the medium produced by the

electromagnetic wave (the Lorentz theory), P standing for the electric moment per unit volume. The question whether Lorentz polarisation should be included in the expression for the refractive index for radio waves was subject to considerable discussions [10] in the past years and appeared to be decided finally in favour of the Sellmeyer's theory as a result of a theoretical analysis by Darwin [11]. But some controversies have, however, arisen in the interpretation of the recent experiments [12] carried out to investigate the way in which the echo-doubling, obtained with the radio waves of frequencies less than that of the gyrofrequency, varies with the frequencies; one school advocating in favour of the Lorentz theory while other the Sellmeyer's theory. A critical examination of this question will be the subject matter of this section and an attempt will be made to analyse the conditions under which such experiments may lead to a definite solution as to the correctness of either of the theories.

When the Lorentz polarisation term is present the refractive index without damping is given by [12]

$$1-\mu^2 = \frac{x_0}{1+\beta x_0 - \frac{1}{2}\frac{y_T^2}{1-x_0+\beta x_0} \mp \frac{1}{2(1-x_0+\beta x_0)}\sqrt{y_T^4+4y_L^2(1-x_0+\beta x_0)^2}} \quad .. \quad (35)$$

whence we obtain for the conditions of total reflection

$$1+\beta x_0 - \frac{1}{2}\frac{y_T^2}{1-x_0+\beta x_0} \mp \frac{1}{2(1-x_0+\beta x_0)}\sqrt{y_T^4+4y_L^2(1-x_0+\beta x_0)^2} = x_0 \quad .. \quad (36)$$

$$= 0 \quad .. \quad (37)$$

according as $\mu = 0$ or $\mu \sim \infty$.

Solving (36) we obtain

either $1-x_0+\beta x_0 = 0$ .. .. .. (38)

or $1-x_0+\beta x_0 = \pm y$ .. .. .. (39)

from which follows

$$\omega_C = 2\pi\nu_C = \sqrt{\frac{4\pi Ne^2}{m}(1-\beta)} \quad .. \quad .. \quad .. \quad (40)$$

$$\omega_C = 2\pi\nu_{C\pm} = \sqrt{\frac{4\pi Ne^2}{m}(1-\beta)+\left(\frac{\omega_H}{2}\right)^2} \pm \frac{\omega_H}{2} \quad .. \quad (41)$$

The relations between the electron density and the penetration frequency being given by

$$N = \frac{\pi m}{(1-\beta)e^2}\nu_C^2 \quad .. \quad .. \quad .. \quad (42)$$

$$N = \frac{\pi m}{(1-\beta)e^2}\nu_C^2\left(1 \pm \frac{\nu_H}{\nu_C}\right) \quad .. \quad .. \quad (43)$$

The results in this case are similar to those as given by $\beta = 0$, the points of total reflection being only shifted by the amount $1-\beta$. Much more important is, however, the reflection for $\mu \sim \infty$. From (37) we obtain a rather complicated cubic equation in x_0, the real positive root of which will give us the required relation between the electron density and the critical penetration frequency of the wave.

We have, namely

$$x_0^3\beta^2(\beta-1)+x_0^2(3\beta^2-2\beta) + x_0(3\beta-1-\beta y^2+y_L^2)+(1-y^2) = 0 \quad .. \quad (44)$$

Taking $\beta = \frac{1}{3}$ it is reduced to

$$x_0^3+\tfrac{9}{2}x_0^2+\tfrac{27}{2}x_0\left(\frac{y^2}{3}-y_L^2\right)+\tfrac{27}{2}(y^2-1) = 0 \quad .. \quad (45)$$

The number of roots which depend on the relative values of y^2 and y_L^2 can be read from the following diagram due to Tayler [13]. For comparison we have also drawn to the same scale the corresponding curves for the case $\beta = 0$ represented by the regions *OMZ* and *XMN*. The curve *ABCDE* in the (y^2, y_L^2) plane is given by the cubic equation

$$8(\tfrac{1}{3}y^2-y_L^2)^3-\tfrac{1}{3}y^4+14y^2y_L^2-3y_L^4-12y^2 = 0.$$

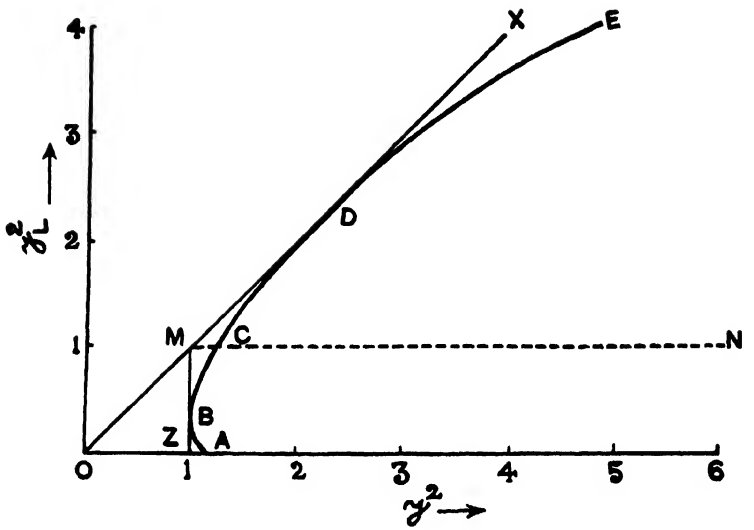


FIG. 4. Ranges of magnetic field and frequency showing the occurrence of infinities in the dispersion curves (μ^2, N) both for $\beta = 0$ and $\beta = \frac{1}{3}$. For $\beta = 0$, x_0 is real and positive in the regions OMZ and XMN. For $\beta = \frac{1}{3}$, x_0 is real and positive between the curves OX and $ZBCDE$.

It is tangential to $y^2 = 1$ at B where $y_L^2 = \frac{1}{3}$ and to the line $y_L^2 = y^2$ at D where $y^2 = y_L^2 = \frac{9}{4}$. We observe the following important results:—

Region.	Range.	No. of infinities in μ^2 for extraordinary wave.		No. of infinities in μ^2 for ordinary wave.	
		$\beta = 0$	$\beta = \frac{1}{3}$	$\beta = 0$	$\beta = \frac{1}{3}$
OMZ ..	$y^2<1$, $y_L^2<1$	1	1	×	×
BMC ..	$y^2>1$, $y_L^2<1$	×	2	×	×
MCD ..	$y^2>1$, $y_L^2>1$	1	2	×	×
EDX ..	$y^2>1$, $y_L^2>1$	1	×	×	2
ECN ..	$y^2>1$, $y_L^2>1$	×	×	1	×

We therefore find that in order to decide whether the Lorentz term should be included in the dispersion equation of

radio waves the region $EDCN$ ($y^2>1$, $y_L^2>1$) will be most suitable for the experimental investigation. We should obtain in this case three reflections of the radio waves in the upper atmosphere, two for ordinary ($\mu=0$, $\mu=\infty$) and one for the extraordinary ($\mu=0$) waves if the Sellmeyer's theory is correct; according to the Lorentz theory on the other hand only two reflections (one ordinary, $\mu=0$ and one extraordinary, $\mu=0$) will be possible. We illustrate this in reference to the conditions at Allahabad for which the relevant datas are as follows:—

$H = \cdot 5182$ Gauss. dip $= 46°$

	$10^{-6}\omega_H$	$10^{-6}\omega_L$	$10^{-6}\nu_H$	$10^{-6}\nu_L$
Ground	9·124	6,563	1,452	1,044
100 km.	8,694	6,253	1,384	·995
250 km.	8·048	5·789	1,280	·921

A typical dispersion curve is shown in fig. 5 drawn for

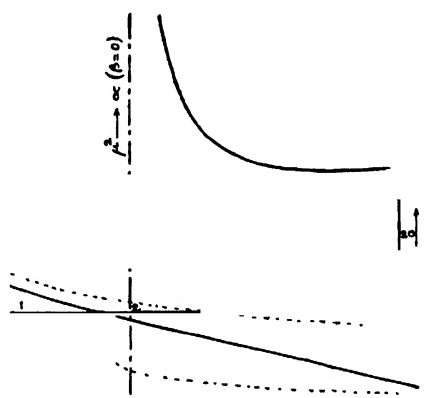


FIG. 5. Comparison of dispersion curves (μ^2, N) with $\beta = 0$ and $\beta = \frac{1}{2}$.
$\lambda = 428{\cdot}4\ m$; $y = \dfrac{eH}{2\pi mc\nu} = 1{\cdot}83$; $\theta = 44°$; $\Delta = 0$.
Curve ——— for $\beta = 0$; Curve ------- for $\beta = \frac{1}{2}$.

$\lambda = 428{\cdot}4\,m$ ($\nu = {\cdot}7$ Mc/sec). We obtain further the electron concentrations which are required to reflect the wave

	$\beta = 0$	$\beta = \frac{1}{3}$	
$10^{-4}N$ =	·611	·917	(ordinary).
$10^{-4}N$ =	1,73	2·60	(extraordinary).
$10^{-4}N$ =	1,97	no reflection	(ordinary).

If we therefore study the echo splitting by the group retardation method with a wave of frequency ·7 Mc/sec onward we should expect, if Sellmeyer's theory is correct, the following results: The waves separate as the frequency is increased and the equivalent height for the component of the ordinary waves given by (31) shall remain nearly constant with the increase of frequency except near the penetration frequency, while for the other component (33) it will rapidly increase, tending asymptotically to infinity at a frequency which is equal to ν_L, i.e. ·92 Mc/sec, because then the ionisation necessary to reflect the wave would be infinite. The equivalent height for the extraordinary component of the wave (32) will, however, also rapidly increase, until the trace representing the height disappears at a frequency which is equal to the gyrofrequency ν_H, i.e. 1,28 Mc/sec, the absorption of the extraordinary wave being very considerable in this case. Whereas on the other hand, if the Lorentz theory is correct, we should obtain two waves, the equivalent height for the ordinary one, as before, remaining constant with the increase of frequency and for the extraordinary wave it increases and then disappears at a frequency, which is equal to the Lorentz frequency and which is much below the gyrofrequency and depends on the concentration of the ions.

The disappearance of the trace of the wave if occurs at a frequency ·92 Mc/sec at Allahabad will therefore indicate the correctness of the Sellmeyer's theory even without measuring the polarisation of the wave. It cannot be the extraordinary wave (Appleton and others), because in that case it would disappear at a frequency 1,28 Mc/sec. Nor can it be interpreted in terms of the Lorentz theory (Booker and Berkner), by assuming that the absorption takes place in the lower part of the *E*-region

where due to the presence of considerable number of ions in excess of the electrons the Lorentz frequency, at which the wave disappears, is much increased. Because this explanation would then require the number of ions, necessary to bring down the Lorentz frequency to a value of ·92 Mc/sec, to be considerably different from that of Washington at the same height of about 100 km. and would not fit into the datas as obtained from the other considerations, such as, to explain the diurnal variation of the terrestrial magnetism.

The confusion which is arisen in the interpretation of the experiments carried out in Sydney, Cambridge and Washington is due to the small dip angle of these places. Because when the dip angle is small, ν_L is very nearly equal to ν_H and the difference between (31) and (33) becomes very difficult to realise in practice. The echo doubling is then practically equivalent to that of the ordinary and extraordinary waves. The polarisation measurement of the echo will evidently not be in a position to decide unambiguously the correctness of either of these two theories; it will rather show a complex character, perhaps a mixture of both the ordinary and the extraordinary waves. For a clear-cut decision of this question we must, however, study the phenomena with wave of frequency which is less than the gyrofrequency and at a place where the dip angle is relatively large so as to make an appreciable difference between ν_L and ν_H. The conditions at Allahabad are quite favourable for this and an investigation of this important problem will be awaited with interest.

Discussion.

We have attempted in the last few pages to investigate the criterion for total reflection directly from its definition in terms of the propagation of energy associated with the electric and magnetic field of the electromagnetic wave. It is to be noticed that the conditions formulated above are identical with those derived on the assumption that the total reflection takes place when the group velocity of the wave train vanishes. This agreement is obvious because both of these interpretations are

merely the statement of the fact, that in case of total reflection no energy flows across the boundary surface of separation of the two mediæ. We have, however, preferred to put the criterion in terms of refractive index because this is easily understood and mathematically much simpler. The difference between the criterion for total reflection $\mu = 0$ and $\mu \sim \infty$ is manifested in the fact, as mentioned before, that in case of the former the equivalent height is greater than the true height whereas for the latter the reverse is the case. It will be therefore interesting to investigate the relation between the true and the equivalent height in these cases for a better understanding of the problem. It will be also interesting to investigate the dispersion in the region of long wavelength at a place where the dip angle is sufficiently large, because this will decide the important question whether we should take account of Lorentz polarisation term in our ionospheric theory or not.

REFERENCES.

[1] R. C. Majumdar.—Zs. f. Phys., Vol. 107, p. 599, 1937; Trans. Bose Res. Inst., Vol. XI, p. 177, 1935-1936; Ind. Jour. Phys., Vol. 12, p. 75, 1938.

[2] E. A. V. Appleton.—Proc. Phys. Soc., Vol. 37, p. 16, 1925; U.R.S.I. Reports (Washington, 1927); Jour. Inst. Elec. Eng., Vol. 71, p. 642, 1932.

D. R. Hartree.—Proc. Camb. Phil. Soc., Vol. 27, p. 143, 1931.

[3] E. A. V. Appleton.—Report on Progress in Physics, Vol. II, p. 129, 1935.

[4] B. D. Pant and R. R. Bajpai, Science and Culture, Vol. 2, p. 409, 1937.

[5] G. Goubau.—Hoch-freq. tech u. Elek. akus., Vol. 45, p. 179, 1935.

[6] R. N. Rai.—Proc. Nat. Inst. Sci. India, Vol. 3, p. 307, 1937.

[7] S. N. Bose.—Ind. Jour. Phys., Vol. 12, p. 121, 1938.

[8] M. Born.—Optik, chapter V, 1933.

[9] P. O. Pedersen.—The propagation of radiowaves, chapter X, 1927.

[10] D. F. Martyn and G. H. Munro.—Nature, Vol. 141, p. 159, 1938.

E. A. V. Appleton, F. T. Farmer and J. A. Ratcliffe.—Nature, Vol. 141, p. 409, 1938.

H. G. Booker and L. V. Berkner.—Nature, Vol. 141, p. 562, 1938.

[11] C. G. Darwin.—Proc. Roy. Soc. Lond. (A), Vol. 146, p. 17, 1934.

[12] R. C. Majumdar.—Trans. Bose Res. Inst., Vol. XI, p. 177, 1935-1936.

[13] M. Tayler.—Proc. Phys. Soc., Vol. 45, p. 245, 1933.

[14] H. G. Booker and L. V. Berkner.—*Loc. cit.*

Published by A. N. Mitter, Superintendent, Bose Institute, 93 Upper Circular Road, Calcutta, and Printed by P. Knight at the Baptist Mission Press, 41A Lower Circular Road, Calcutta.